

Moisture loss studies in nectarines
(*Prunus persica* var. *nectarina*)

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DECLARATION

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SUMMARY

Moisture loss studies in nectarines (*Prunus persica* var. *nectarina*)

Moisture loss during long term storage is one of the main post-harvest problems in nectarines. The long handling chain to which fruit are exposed to, from harvest until the end of shelf-life, exposes fruit to moisture loss. Moisture loss occurs as a result of the vapour pressure deficit (VPD) between the fruit and surrounding atmosphere. In addition to causing loss of saleable weight, moisture loss also results in fruit having a shrivelled appearance. Moisture is lost from fruit through various openings in the fruit peel such as micro-cracks and lenticels.

In this study we investigated the effect of fruit to fruit variation, harvest date, tree and orchard effects and cultivar differences on the variation in water vapour permeance ($P'H_2O$) of three nectarine cultivars namely 'Alpine', 'Summer Bright' and 'August Red'. The study showed that large fruit to fruit differences were the main contributor (>45%) to the variation in $P'H_2O$, followed by harvest date (>35%), cultivar differences (>7%) and orchard effects (>3%) whilst tree effects did not contribute to $P'H_2O$. Generally, the $P'H_2O$ of all three cultivars increased steadily as the harvest date approached and continued to increase post-harvest, but $P'H_2O$ at optimum harvest was not closely correlated to their susceptibility to shrivel.

In addition, 'August Red' nectarines were exposed to different handling chains from harvest until the end of shelf-life to determine the VPD at different stages in the handling chain in order to establish the point which is most effective in reducing moisture loss and shrivel. The results indicated that none of the proposed handling chains performed better than the current standard handling protocol in reducing moisture loss and shrivel. This protocol stipulates that nectarines should be harvested during the cooler time of the day and field heat should be removed as soon as possible after harvesting. Furthermore, the handling protocol requires that nectarines should be packed within 12 hours of arrival at the pack-house.

Several researchers have reported that silicon containing fertilizers improve fruit quality and we therefore also investigated whether pre-harvest applications of potassium silicate (K_2SiO_3) can reduce post-harvest moisture loss, shrivel and split pit in 'Southern Glo'

nectarines. The results showed that both soil and foliar K_2SiO_3 applications were not effective in reducing post-harvest moisture loss, shrivel or the incidence of split pit in 'Southern Glo' nectarines. For future studies, it is recommended to increase the frequency of K_2SiO_3 applications.

The study also looked at the effectiveness of different packaging films in reducing moisture loss and shrivel in 'August Red' and 'Alpine' nectarines. Failure to package fruit optimally may result in weight loss, shrivel, decay and the incidence of internal defects such as woolliness, pulpiness and over-ripeness. The results showed that the use of Xtend® and high density poly-ethylene (HDPE) bags significantly reduced moisture loss and shrivel in nectarines in both pulp trays and plastic punnets. The standard nectarine HDPE wrappers resulted in significantly higher percentage mass loss as well as shrivel incidence in 'Alpine' nectarines.

It is therefore important to reduce moisture loss at harvest by following the standard handling protocol and by packing fruit optimally.

OPSOMMING

Studies oor vogverlies in nektarienvrugte (*Prunus persica* var. *nectarina*)

Vogverlies tydens langtermynopberging is een van die hoof na-oes probleme wat nektariens ervaar. Vrugte word aan 'n lang hanteringsketting blootgestel vanaf oes tot die einde van rակlewe en dit lei tot vogverlies. Vogverlies vind plaas as gevolg van die water dampdruk tekort (WDDT) tussen die vrug en die omringende atmosfeer. Buiten dat dit die verkoopbare gewig van vrugte verminder, sal vogverlies ook daartoe lei dat vrugte 'n verrimpelde voorkoms het. Vog gaan verlore uit die vrug deur verskeie openinge in die skil byvoorbeeld mikrokrakies en lentiselle.

In hierdie studie het ons die effek van vrug tot vrug variasie, oesdatum, boom- en boord effekte en kultivarverskille op vogdeurlaatbaarheid ($P'H_2O$) van drie nektarienkultivars, naamlik 'Alpine', 'Summer Bright' en 'August Red' ondersoek. Die studie het getoon dat vrug tot vrug variasie die hoof bydrae (>45%) tot verskille in $P'H_2O$ gemaak het, gevolg deur oesdatum (>35%), kultivar verskille (>7%) en boord effekte (>3%) terwyl boom effekte geen bydrae gelewer het tot $P'H_2O$ nie. Oor die algemeen het die $P'H_2O$ van al drie kultivars geleidelik gestyg soos die optimum oesdatum nader gekom het en het verder gestyg na die optimum oesdatum. Die $P'H_2O$ tydens optimum oes was egter nie goed gekorreleer met die kultivar se geneigdheid om te verrimpel nie.

Verder is 'August Red' nektariens blootgestel aan verskillende hanteringsprotokolle vanaf oes tot na rակlewe om die WDDT te bepaal tydens verskillende tydstippe in die hanteringsketting om te bepaal tydens watter periode die WDDT die beste gereduseer kon word om sodoende vogverlies en verrimpeling te verminder. Nie een van die voorgestelde hanteringskettings het beter presteer om vogverlies en verrimpeling te verminder as die standaard, aanbevole hanteringsketting nie. Hierdie protokol stipuleer dat nektariens gedurende die koeler tyd van die dag geoes moet word en dat veldhitte so vinnig moontlik na oes verwyder moet word. Verder vereis die protokol dat nektariens binne 12 uur na aankoms by die pakhuis verpak moet word.

Verskeie navorsers het aangedui dat silikonbevattende kunsmisstowwe vrugkwaliteit kan verbeter. Ons het dus ondersoek of voor-oes toedienings van kaliumsilikaat (K_2SiO_3) na-oes vogverlies, verrimpeling en gebreekte pitte in 'Southern Glo' nektariens kan

verminder. Beide grond- en blaartoedienings van K_2SiO_3 was oneffektief om na-oes vogverlies, verrimpeling en gebreekte pitte in 'Southern Glo' nektariens te verminder. In toekomstige studies moet die frekwensie van K_2SiO_3 toedienings dalk verhoog word.

Hierdie studie het ook die effektiwiteit van verskillende verpakkingsmateriale om vogverlies en verrimpeling te verminder op 'August Red' en 'Alpine' nektariens ondersoek. Wanneer vrugte nie optimaal verpak word nie kan massaverlies, verrimpeling, verrotting en interne kwaliteitsverliese soos voosheid, pulpagtigheid en oorrypheid voorkom. Resultate het daarop gedui dat die gebruik van Xtend® en hoë digtheid poli-etileen (HDPE) sakke vogverlies en verrimpeling betekenisvol kan verminder in nektariens in beide pulprakkies en plastiek houers. Die standaard nektarien HDPE "wrappers" het tot betekenisvol meer massa verlies en die voorkoms van verrimpeling gelei in 'Alpine' nektariens.

Dit is dus belangrik om vogverlies naoes te verminder deur die standaard hanteringsprotokol te volg en nektariens optimaal te verpak.

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NOTE

This thesis is a compilation of chapters, starting with a literature review, followed by four research papers. Each paper is prepared as a scientific paper for submission to *Postharvest Biology and Technology*. Repetition or duplication between papers might therefore be necessary.

GENERAL INTRODUCTION AND OBJECTIVES

The export of nectarines can be very challenging since fruit usually spend about four weeks in cold storage during loading, accumulation of containers and shipment to overseas market (Laubscher, 2006). Moisture loss is one of the main post-harvest problems that affect the quality of peaches and nectarines during long term storage (Crisosto and Day, 2012). To ensure optimum post-harvest quality, stone fruit such as peach and nectarine should be protected from excessive post-harvest moisture loss (Crisosto and Day, 2012). According to Holcroft (2015) peaches and nectarines have a water content of approximately 89% and will show symptoms of shrivel when losing 19% or more of this water. Moisture loss during long term storage can result in fruit having a shrivelled appearance rendering them unsaleable (Maguire et al., 2000). In addition to moisture loss and shrivel, other problems which are associated with long periods of storage are decay and the incidence of internal defects such as woolliness, pulpiness and over-ripeness (Aharoni et al., 2007; Kaur et al., 2013; Porat et al., 2009).

In order to gain a better understanding of moisture loss in nectarines and how it can be ameliorated, a literature review was done followed by a number of trials. According to Maguire et al. (2001), the fruit cuticle modulates loss of moisture from the fruit and the efficacy of the cuticle to reduce moisture loss depends on its composition and structure. The ease with which water vapour can escape from a fruit is called the water vapour permeance ($P'H_2O$) (Maguire et al., 2000). In a constant environment, the $P'H_2O$ of a fruit surface can be calculated from the rate of water loss using Fick's first law of diffusion (Maguire et al., 1999). The composition and structure of the cuticle varies from fruit to fruit depending on the cultivar, harvest maturity, orchard and tree effects, as well as growing conditions (Lescourret et al., 2001; Maguire et al., 2000; Theron, 2015). A study by Maguire et al. (2000) quantified the contribution of each of these factors to the total variation that is observed in the $P'H_2O$ of apple fruit. Theron (2015) also quantified the contribution of each of these factors to the total variation observed in Japanese plums. Currently no information exists on the $P'H_2O$ of nectarines produced in South Africa. Therefore, in Paper 1 we determined the $P'H_2O$ of 'Alpine' (susceptible to postharvest moisture loss), 'Summer Bright' (not susceptible to postharvest moisture loss) and

‘August Red’ (highly susceptible to postharvest moisture loss) nectarines. The research also aimed to establish if fruit to fruit differences, cultivar differences, harvest maturity, orchard and tree effects have an effect on the P_{H₂O} of these nectarine cultivars.

The South African fruit export handling chains consist of many steps and role-players from harvest until fruit reaches the consumers (Goedhals-Gerber et al., 2015). These long handling chains expose nectarines to post-harvest moisture loss and may affect their quality when they finally reach their markets. At harvest, fruit have a fresh appearance and crisp texture. However, harvesting removes the fruit from its water supply and the fruit will begin to lose moisture without replenishing its moisture content (Goedhals-Gerber et al., 2015; Holcroft, 2015). Currently the handling protocol for nectarines in South Africa is to harvest fruit during the cooler time of the day (temperature under 25 °C) and to remove field heat as soon as possible after harvesting by cooling fruit to just above the dew point temperature of the pack-house (HORTGRO, 2014). In addition, the handling protocol requires that nectarines should be packed within 12 hrs of arrival at the pack-house (HORTGRO, 2014). Low temperature disorders such as woolliness and pulpiness (internal breakdown) limit the cold-storage life of nectarines and reduce the quality of nectarines in the markets (Lurie and Crisosto, 2005). Pre-ripening of nectarines has been done for over 50 years and involves a delay in the commencement of cooling by keeping fruit at 20 °C for approximately 48 hours after harvest (Laubscher, 2006; Nanos and Mitchell, 1991). Therefore, in Paper 2 we determined the vapour pressure deficit (VPD) between ‘August Red’ nectarines and their environment during different simulated post-harvest handling chains. This information will show where in the handling chain the risk for moisture loss is the highest, and with this information at hand the industry will be better equipped to create and apply optimum handling protocols to prevent moisture loss.

The use of silicon containing fertilizers to improve the quality of fruit has been investigated by several researchers (Mditshwa et al., 2013; Stamatakis et al., 2003; Tarabih et al., 2014). Silicon is deposited in the plant cell walls and this helps to reinforce the cell walls by interacting with cell wall pectins and polyphenols (Stamatakis et al., 2003). This assists in protecting the plant from various stresses and disease causing

pathogens (Epstein, 1999; Stamatakis et al., 2003). Silicon containing fertilizers reduced post-harvest weight loss in citrus fruit (Mditshwa et al., 2013) and ‘Anna’ apples (Mditshwa et al., 2013; Tarabih et al., 2014). The application of silicon containing fertilizers may be a possible solution to post-harvest moisture loss in nectarines and therefore in Paper 3 we investigated if pre-harvest K_2SiO_3 applications can maintain fruit quality post-harvest while reducing the incidence of shrivel and split pit in ‘Southern Glo’ nectarines.

Lastly, in Paper 4 we compared the effectiveness of different packaging materials in reducing moisture loss, shrivel incidence, decay and internal breakdown (pulpiness, woolliness and over-ripeness) in nectarines and also established whether or not different types of packaging material should be used for large (± 65 mm diameter) and small (± 56 mm diameter) nectarines. The long storage duration during shipment exposes fruit to loss of quality and in order to minimize this, it is important that proper packaging materials are used. Failure to package fruit optimally may result in moisture loss, shrivel, decay and the incidence of internal defects such as woolliness, pulpiness and over-ripeness (Aharoni et al., 2007; Kaur et al., 2013; Porat et al., 2009). The packaging materials used as treatments in this trial were the standard nectarine wrapper (HDPE), Nectarine Xtend[®] bag, HDPE bag (54 x 2 mm perforations) and the HDPE bag (34 x 4 mm perforations). ‘August Red’ which is susceptible to shrivel was used in the 2014/2015 season, whilst ‘Alpine’ which is also susceptible to shrivel was used in the 2015/2016 season.

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LITERATURE REVIEW:

Post-harvest moisture loss in nectarines (*Prunus persica* var. *nectarina*) and measures that can be taken to reduce this problem

1. Introduction

Nectarine (*Prunus persica* var. *nectarina*) belongs to the Rosaceae family and is native to China where it has been cultivated for over 2000 years (Uthairatanakij, 2004). Nectarines are closely related to peaches, the difference between them is that nectarines lack the pubescence that is found on peaches (Layne and Bassi, 2008) due to a recessive gene found in nectarines (Uthairatanakij, 2004). In South Africa, nectarines are mainly grown in the Western Cape region (HORTGRO, 2014). The major production areas in the Western Cape are Ceres (902 hectares), Wolseley / Tulbagh (286 hectares) and Paarl (230 hectares) (HORTGRO, 2014). Most of the nectarines produced in South Africa are exported, and in the 2013/2014 season the major export destination for nectarines were the United Kingdom (54%), Europe and Russia (22%) and the Middle East (19%) (HORTGRO, 2014).

Moisture loss is one of the main post-harvest problems that affect the quality of peaches and nectarines during long term storage (Crisosto and Day, 2012). To ensure optimum post-harvest life, stone fruit such as peaches and nectarines should be protected from excessive post-harvest moisture loss (Crisosto and Day, 2012). According to Holcroft (2015) peaches and nectarines have a water content of about 89% and will only show symptoms of shrivel when they have lost at least 19% of this water. Moisture loss during long term storage can result in fruit with a shrivelled appearance rendering them unsaleable (Maguire et al., 2000). As a result of moisture loss, there is loss in saleable weight as well as deterioration of fruit quality (Sastry, 1985). Moisture is lost from the fruit as a result of the vapour pressure deficit (VPD) between the fruit and the surrounding atmosphere. Fruit moisture loss occurs through various parts of the fruit surface, these include the stomata, lenticels, cuticle, and epicuticular wax platelets (Díaz-Pérez et al.,

2007). Moisture loss accounts for over 97% of the total weight loss in harvested produce (Díaz-Pérez et al., 2007).

The purpose of this review is to look at the factors that affect post-harvest moisture loss in nectarines as well as look at the measures that can be implemented to mitigate the effects of moisture loss. An important strategy for minimizing post-harvest moisture loss is maintaining low temperatures during post-harvest handling and storage (Paull, 1999; Henriad et al., 2005). A combination of low temperature and high humidity during storage will reduce the VPD between the fruit and the storage atmosphere and this results in reduced moisture loss (Paull, 1999). Avoiding unnecessary delays in pre-cooling of fruit will also help to reduce post-harvest moisture loss (Crisosto and Valero, 2008). Strategies to further reduce moisture loss include the use of modified atmosphere packaging (MAP) and application of edible fruit coatings (Dhall, 2013; Nasr et al., 2013).

2. The nectarine fruit

2.1. Taxonomy and origin

As mentioned earlier, nectarine (*Prunus persica* var. *nectarina*) belongs to the Rosaceae family and is closely related and genetically similar to peach, the occurrence of one recessive gene in nectarines is responsible for the lack of trichomes (pubescence) (Uthairatanakij, 2004). Because of this, nectarines have a smooth peel lacking epidermal hairs. According to Layne and Bassi (2008), the smooth peel of nectarines makes them more susceptible to mechanical and pest damage compared to peaches. Both peaches and nectarines have a limited genetic base due to the low number of genotypes used as parents in breeding programs (Yoon et al., 2006).

According to Hummer and Janick (2009), there are basically two types of flesh texture in nectarines, namely melting and non-melting. The texture for the melting flesh softens during the last stage of ripening in response to increased ethylene production, the non-melting types remain firm even during ripening and show a little softening when they are overripe (Ghiani et al., 2011). Growers should benefit more from the non-melting cultivars because of less damage during harvest, transport and storage (Reid et al.,

2006). Nectarines are also classified as either cling- or freestone (Uthairatanakij, 2004). In freestone cultivars the stone does not strongly adhere to the flesh as in clingstone cultivars (Uthairatanakij, 2004).

2.2. Structure of the nectarine fruit

Nectarines are drupes, and have a thin outer layer (peel; epicarp), edible flesh beneath this layer (fleshy mesocarp) and a hard lignified stone or wall (endocarp) in the centre of the fruit (Kader and Mitchell, 1989). The epicarp acts as a protective layer and is composed of epidermal and hypodermal cells (Uthairatanakij, 2004). The cuticle is composed of wax and serves to reduce moisture loss and also reduces entry of pathogens into the fruit (Kader and Mitchell, 1989). Most of the mechanical strength of the peel is a result of the heavy-walled epidermal cells. The mesocarp is the main edible portion of nectarines and consists of parenchyma cells which have thin cell walls as well as high water content (Uthairatanakij, 2004).

2.3. Nectarine fruit ripening

During ripening the fruit softens and this is important as it improves the sensory quality of the fruit (Heyes and Townsend, 1992). Ripening and fruit softening is a result of the breakdown of cellulose and pectins found in the cell walls (Payasi et al., 2009). During ripening there are changes to the structure and composition of various components of the fruit i.e. carbohydrates, phenols, lipids and volatile compounds (Uthairatanakij, 2004). Nectarines are usually harvested when they are mature but before ripening starts (Kader and Mitchell, 1989). Peaches and nectarines are climacteric fruit, they show a rise in ethylene production during ripening. This ethylene production is important during the ripening process, and a concentration of between 1-3 ppm will initiate ripening (Kader and Mitchell, 1989).

Nectarines are stored at low temperature (usually -0.5 °C) to delay the ripening process. The fruit will start to ripen and decrease in firmness when they are taken out of cold storage. Nectarines contain some cell wall degrading enzymes which increase in activity during ripening, examples of such enzymes are cellulase, polygalacturonase (PG)

and exopolygalacturonase (Heyes and Townsend, 1992). According to Heyes and Townsend (1992) softening is also a result of increased proton pumping across the plasma membrane, this lowers the pH and loosen acid-labile bonds. This is facilitated by the enzyme plasma membrane H^+ -ATPase. The lowering of pH also activates other cell wall degrading enzymes such as endo-PG (Heyes and Townsend, 1992).

3. Post-harvest factors affecting moisture loss of fruit

3.1. Driving force for moisture loss (vapour pressure deficit)

Water loss in harvested fruit involves the movement of water vapour down a concentration gradient from the fruit surface to the surrounding environment (Maguire, 1998). The rate at which the fruit will lose water varies directly with the VPD between the fruit and the surrounding atmosphere (Whitelock et al., 1994). VPD is the driving force for moisture loss and it describes the difference between the water vapour partial pressure in the fruit and in the surrounding atmosphere (Toivonen and Hodges, 2005). The driving force for moisture loss from fruit can be described in terms of vapour pressure, concentration of moisture or differences in water activity across membranes (Veraverbeke et al., 2003b). The water vapour partial pressure inside the fruit is considered to be almost saturated (Paull, 1999). In most storage environments, the partial pressure of the air is below saturation level and the result is a net movement of water vapour from the fruit into the surrounding environment (Maguire, 1998).

Soon after harvest, the produce usually has a lot of field heat and placing the produce in cold storage without removing the field heat will increase the driving force for moisture loss (Paull, 1999; Toivonen and Hodges, 2005). It is therefore very important to cool the produce as soon as possible after harvest so that moisture loss is reduced (Toivonen and Hodges, 2005). Cooling mechanisms used in nectarines include forced air cooling (FAC), hydro-cooling and room cooling (Kalbasi-ashtari, 2004). (Whitelock et al., 1994) found that weight loss in peaches varied directly with the VPD. It is therefore important to implement measures to reduce the VPD during storage, such as reducing storage temperature, high relative humidity (RH) and reduced air velocity.

3.2. Fruit surface temperature

Fruit surface temperature is a major determinant of moisture loss from stored produce (Maguire, 1998). Heat moves through conduction from inside the fruit to the surface of the fruit, this heat is then transferred from the fruit surface to the surrounding atmosphere causing fruit to lose moisture. Fruit will continue to respire even after they are harvested, respiration produces heat and this heat accumulates within the fruit (Burg, 2004). Accumulation of respiratory heat increases the water vapour partial pressure within the fruit and this in turn increases the driving force for moisture loss (Maguire et al. 2000). By storing produce at low temperatures, the rate of respiration is reduced. A reduction in respiration rate will reduce the level of respiratory heat, this reduces the water vapour pressure of the fruit leading to reduced moisture loss (Becker and Fricke, 1996).

3.3. Relative humidity of storage atmosphere

The success of the fruit export industry depends on the ability to provide the market with high quality products (Whitelock et al., 1994). Nectarines are usually in storage for four weeks or longer during transit to their markets, therefore if storage conditions are not ideal a lot of moisture can be lost during this period (Paull, 1999), it is therefore important to minimize moisture loss during this time. Relative humidity of the storage atmosphere is one of the factors that affect moisture loss (Maguire et al., 2001). According to Paull (1999), RH is affected by the surface area of the evaporation coil in the storage unit as well as the difference in temperature between the air and the coil. The challenge with maintaining a high RH in storage units is that any small fluctuations in temperature will cause considerable changes in the RH. However, standard technologies which are being used make it easier to maintain the humidity in storage units at acceptable levels (Paull, 1999). Water loss from stored produce occurs when the RH in the cooling room is below the humidity inside the fruit, the humidity in the fruit is considered to be 100% (Veraverbeke et al., 2003b).

At high RH the VPD between the produce surface and the storage atmosphere is reduced and this will reduce moisture loss from the produce (Whitelock et al., 1994). High

RH during storage can reduce cuticular water loss especially when it is coupled with low temperature and low air velocities (Henriod, 2006). Brusewitz et al. (1992) found that high RH was indeed beneficial in maintaining high quality in stored peaches. Paull (1999) and Mitchell & Crisosto (1995) also support these findings and found that peaches and nectarines should be stored at a RH of 95% for optimum quality. Although high RH is beneficial in reducing moisture loss, very high RH (> 95%) can lead to growth of bacteria, fungi and other pathogens (Wu, 2010).

3.4. Storage Temperature

Storage temperature is one of the important factors that affect moisture loss in many fruit types and nectarines are no exception. Temperature regulates the rate of most physiological and biochemical processes which occur within the fruit (Khorshidi et al., 2010) and maintaining low temperatures during storage is a key factor in extending post-harvest life and quality of stored produce (Henriod et al., 2005). The storage environment can be easily saturated with water vapour when the temperature is lower (Wijewardane and Guleria, 2013). Optimum storage temperatures differ among produce and cultivars. For nectarines the optimum storage temperature is -0.5 °C (Paull, 1999). At high storage temperatures the rate of respiration of the produce will increase and this will lead to increased weight loss. An increase in storage temperature is likely to increase the VPD between the fruit and the storage atmosphere and this will subsequently lead to increased moisture loss from the fruit (Paull, 1999).

Incidence of internal disorders will also increase as the storage temperature increases. According to Paull (1999) woolliness and mesocarp browning will be more prevalent in nectarines stored at temperatures of between 2 °C and 5 °C compared to nectarines stored at -0.5 °C. Obenland & Carroll (2000) found that internal breakdown occurs more frequently in nectarines stored between 2.2 °C and 7.7 °C. According to Von Mollendorff et al. (1993), the percentage of nectarines with woolliness was higher in fruit stored at 3 °C for 4 weeks than those stored at -0.5 °C for 4 weeks. In addition to this Von Mollendorff et al. (1993) also found that the activity of polyphenol oxidase in nectarines is inhibited at low temperatures, which lowers the incidence of browning. The storage

potential of 'August Red' and 'Summer Bright' nectarines is five weeks when they are stored at 0 °C, while at 5 °C the storage potential is reduced to only three weeks (Crisosto and Day, 2012).

3.5. Surface area to volume ratio

Rate of moisture loss from stored produce depends on the diffusion of water from inside the fruit to the fruit surface and the evaporation of water from the fruit surface to the surrounding environment (Lownds et al., 1993). According to Lownds et al. (1993), moisture loss from the surface of a fruit is positively correlated to its surface area to volume ratio. Research carried out in bell pepper fruit showed that smaller fruit have a higher surface area to volume ratio and are more sensitive to moisture loss compared to larger fruit which have a smaller surface area (Lownds et al., 1993). This was confirmed by Boonyakiat et al. (2012) who found a larger proportional weight loss in smaller tangerine fruit compared to bigger fruit after being subjected to the same period of shelf-life. Díaz-Pérez et al. (2007) also confirmed this for bell pepper fruit. A high surface area to volume ratio means that the fruit will have a greater diffusional area per unit volume and this results in a higher rate of moisture loss (Díaz-Pérez et al., 2007).

3.6. Air velocity in storage atmosphere

Air velocity is another important factor that affects moisture loss in stored produce. Passing air over the fruit at high speed has both positive and negative impacts (Whitelock et al., 1994). High air velocity is beneficial when cooling the fruit, but once the fruit has been cooled, reducing air speed will reduce moisture loss from the fruit (Whitelock et al., 1994). As the fruit is losing moisture, it creates a boundary layer of high humidity around the fruit and this lowers the VPD between the fruit and the surrounding atmosphere (Sastry, 1985). If the speed of the air passing over the fruit is too high, it can blow away the high RH micro-climate surrounding the fruit and this increases loss of water from the fruit (Sastry, 1985; Mitchell and Crisosto, 1995). Air velocity has a considerable impact on the VPD and resistivity of nectarines to moisture loss (Crisosto et al., 1995; Whitelock et al., 1994). According to Crisosto and Valero (2008) the ideal air velocity during nectarine cold storage is $0.0236 \text{ m}^3 \text{ s}^{-1}$.

3.7. Pre-cooling of produce

Pre-cooling is the process of removing field heat from harvested produce in order to slow down biochemical reactions and reduce evaporative loss of moisture (Brosnan and Sun, 2001; Jiang et al., 2006). Pre-cooling reduces the VPD between the fruit and the surrounding atmosphere (Mitchell and Crisosto, 1995). The effects of high temperature depends on the length of time products are exposed to a certain temperature, unnecessary delays in pre-cooling should therefore be avoided (Crisosto et al., 1995; Brosnan and Sun, 2001). Pre-cooling helps to lower product temperature more rapidly and this helps to reduce incidence of wilting and shrivelling (Wijewardane and Guleria, 2013). Wijewardane and Guleria (2013) found that weight loss was slower in pre-cooled apples compared to apples that were not pre-cooled.

Methods of pre-cooling which can be used in nectarines include forced-air cooling, hydro cooling, room cooling and vacuum cooling (Kalbasi-ashtari, 2004; Jiang et al., 2006). Forced-air cooling is a common method used for nectarines (Mitchell and Crisosto, 1995) and it involves forcing cold air to move over the produce at high speed, this causes transfer of heat from the fruit to the cold air (Mitchell and Crisosto, 1995; Kalbasi-ashtari, 2004). Although this method is effective, it is expensive and it also leads to loss of surface water and weight loss in the produce (Kalbasi-ashtari, 2004). Hydro-cooling is also recommended in stone fruit such as peaches, nectarines and cherries (Thompson and Chen, 1989). It involves immersing or spraying the produce with cold water to reduce their temperature (Becker and Fricke, 1996). This avoids loss of water from the produce and can result in the produce absorbing extra moisture (Kalbasi-ashtari, 2004). According to Mitchell and Crisosto (1995) room cooling is a slower method of cooling stone fruits and has generally been replaced by faster methods of cooling such as forced air cooling. Thompson and Chen (1989) reported that vacuum cooling results in moisture loss of about 1% for every 5-6 °C drop in temperature and this can cause unacceptable loss of quality in the produce.

4. Nectarine cuticle and water vapour permeance

4.1. Structure of the nectarine cuticle

The fruit peel is primarily composed of four layers of tissue namely the hypodermis, epidermis, epidermal hairs and the cuticle (Maguire, 1998). Being the outermost layer of the peel of a mature fruit, the cuticle is the most important barrier against moisture loss in stored fruit and vegetables (Lownds et al., 1993). The number of stomata in peach is determined at anthesis and the density and functionality of stomata decreases as the fruit size increases because as the fruit size increases the number of stomata is diluted on the fruit surface area (Gibert et al., 2010). The stomata in peach therefore lose their functionality during growth and are converted into lenticels (Gibert et al., 2010). During storage of apples there are no functional stomata, therefore lenticels and surface cracks located on the cuticle are more important channels of water loss (Veraverbeke et al., 2003a). The cuticle is therefore the main path through which moisture is lost in a mature fruit and its structure and composition plays a major role in modulating moisture loss (Gibert et al., 2005). The cuticle is a bi-layered membrane consisting of a cutin and wax layer with different diffusion and osmotic properties (Veraverbeke et al. 2003a). According to Riederer and Schreiber (2001) the cuticle is composed of polysaccharides, solvent-soluble lipids as well as fatty acids linked through ester, covalent and electronic bonds. The role of the cuticle is particularly important after harvest because at this stage the fruit will not be receiving any more water from the parent plant (Díaz-Pérez et al., 2007).

Two groups of lipids are present in the cuticle i.e. insoluble polymetric cutins and soluble cuticular lipids of which the soluble cuticular lipids provide the main barrier to reduce moisture loss, however their effectiveness in reducing moisture loss depends on their structure and chemical composition (Maguire et al., 2001). Riederer and Schreiber (2001), found that that water permeance for tomato increased by a factor of 20 when the soluble cuticular lipids were removed from the tomato peel. The permeability of the waxy cuticle is not determined by the cuticle thickness but rather by the chemical composition and arrangement of the cuticle components (Leide et al., 2007). Although the cuticle is important in reducing moisture loss, it should also be able to allow proper gas exchange

to occur so that normal aerobic respiration continues to take place in the stored produce (Maguire et al., 2001). In addition to its role in reducing moisture loss, the cuticle is also involved in the development of cracking and preventing pathogens from invading the fruit (Lara et al., 2014).

4.2. Water conductance through the nectarine cuticle

Water is transported across the cuticle by simple diffusion down a water potential gradient (Riederer and Schreiber, 2001). The water molecules are sorped by the cuticular membrane on one side and desorped on the other side (Riederer and Schreiber, 2001). The cuticle has properties which are similar to those of a solution-diffusion membrane and therefore molecules diffuse through it as individual molecules (Karbulková et al., 2008). Schreiber et al. (2001) suggested that there are two pathways for water movement through the cuticle. The lipid fraction of the cuticle forms the first pathway whereas the second pathway is restricted to hydrated polar groups (-OH and -COOH) (Schreiber et al., 2001).

There are a number of factors that affect the permeability and water movement through the fruit peel, including cultivar, harvest maturity, orchard and tree effects, and growing conditions (Maguire et al., 2000; Lescourret et al., 2001). Lescourret et al. (2001) found that surface conductance of peach increased with fresh fruit mass but the pattern differed with cultivar and fruit-to-fruit variation. The rate of water conductance through the nectarine fruit depends on fruit area and water vapour efflux per unit of area (Lescourret et al., 2001). The water vapour permeance (P'_{H_2O}) can be modelled by Fick's first law of diffusion (Maguire et al. 2000; Veraverbeke et al. 2003b). The permeance (P'_{H_2O}) can be calculated using the following equation (Maguire et al., 2000).

$$P'_{H_2O} = \frac{r'_{H_2O}}{(\Delta p_{H_2O} \times A)}$$

Where:

r'_{H_2O} = rate of moisture loss from the fruit (mol.s^{-1})

A = Area of the fruit (m^2)

Δp_{H_2O} = partial pressure difference between the environment and the inside of the fruit (Pa)

P'_{H_2O} = permeance of the fruit surface to water vapour ($\text{mol.s}^{-1}.\text{m}^{-2}.\text{Pa}^{-1}$)

4.3. Effect of harvesting date on the permeability of the nectarine fruit peel

Little research has been done on the permeability of nectarine fruit peel with respect to harvest date. However, research by Lescourret et al. (2001) in three peach cultivars (Alexandra, Suncrest and Opale) and one nectarine cultivar (Big Top) showed that as the fruit size increases the peel permeability also increased. Therefore as the harvesting date approaches, both the fruit size and peel permeability will increase. Theron (2015) also found that the peel permeability of three Japanese plums ('African Delight™', 'Laetitia' and 'Songold') increased as fruit matured beyond their optimum maturity. He attributed this to changes in cuticle thickness and composition of cuticular waxes. Lescourret et al. (2001) further found that an increase in peel permeability in peaches and nectarines might be due to surface cracks that spread over the surface of the fruit as it grows. Research on apples showed that the peel permeability of four apple cultivars (Braeburn, Pacific Rose™, Granny Smith, and Cripps Pink) also increased steadily as the harvesting date approached (Maguire et al., 2000).

4.4. Role of silicon in reducing moisture loss

Silicon (Si) is an important nutrient involved in protecting plants against a wide range of biotic and abiotic stresses including moisture loss (Epstein, 2009). Silicon is the second most abundant element in the earth's crust (28%) and its abundance might be the reason why it is not considered as one of the essential plant nutrients (Tesfagiorgis and Laing, 2013). Furthermore, Si is the only plant nutrient that is not toxic to the plant even when it is absorbed in excess (Currie and Perry, 2007). Potassium silicate (K_2SiO_3) is the most common source of Si in agriculture (Tarabih et al., 2014). According to Currie and Perry (2007), Si is mainly taken up by plants in the form of soluble silicic acid ($\text{Si}(\text{OH})_4$). Silicon is deposited onto the cell walls of plant cells and this helps to reinforce the cell walls, protecting the plant from various stresses and disease causing pathogens (Epstein,

1999; Stamatakis et al., 2003). Plants which are lacking in Si are usually weak and show abnormal growth patterns (Currie and Perry, 2007).

Silicon helps to reduce the post-harvest moisture loss from fruit and the use of K_2SiO_3 post-harvest dips to ameliorate moisture loss problems in fruits has been reported in lemons by Mditshwa et al. (2013) and apples by Tarabih et al. (2014). Mditshwa et al. (2013) found that post-harvest application of $50 \text{ mg L}^{-1} K_2SiO_3$ significantly reduced weight loss and chilling injury in lemons. However, Mditshwa et al. (2013) highlighted that post-harvest Si dips impaired fruit quality and therefore pre-harvest Si application should be considered as a way to mitigate moisture loss and chilling injury in fruits. Epstein (1999) reported that Si helps to protect plants against toxicity of other elements such as manganese and aluminium. Stamatakis et al. (2003) found that pre-harvest application of Si in tomatoes enhanced the translocation of calcium to both the leaves and the fruit, resulting in better quality fruit.

5. Packaging and fruit coatings to reduce moisture loss

5.1. Modified atmosphere packaging

Modified atmosphere packaging (MAP) has been widely used to maintain post-harvest quality as well as extend the storage life of many fruit including nectarines (Nasr et al., 2013; Cefola et al., 2014). MAP involves the use of micro-perforated polyethylene bags to create an atmosphere of high RH, relatively high CO_2 concentration and low O_2 concentration inside the packaging (An et al., 2007; Singh et al., 2013). During MAP plastic films with different perforations, chemical composition and materials are used (Azene et al., 2014). The atmospheric conditions within the packaging change as a result of respiration of the fruit and the gas diffusion properties of the packaging material (An et al., 2007). The high RH inside the packaging lowers the VPD between the fruit and the surrounding air and this results in fruit losing less moisture (Maguire et al., 2000). Types of MAP vary depending on the way in which they modify the internal RH and gaseous environment and are also designed to fit specific fruit types (Henriod, 2006). Crouch (1998) found that 'Laetitia' plums packed in poly-ethylene $55\mu\text{m}$ and polypropylene P-

Plus 160 bags (modified atmosphere) were less shrivelled compared to fruit not in bags or fruit in paper wrappers. The polymeric film around the fruit prevents the fruit from losing too much moisture and also slows down the ripening of fruits whilst they are still in storage, the rate of fruit ripening will increase when fruit is moved from cold-storage to shelf-life (Singh et al., 2013).

5.2. High density poly-ethylene and low density poly-ethylene

High density poly-ethylene (HDPE) and low density poly-ethylene (LDPE) are common types of packaging materials used during fruit storage (Allahvaisi, 2012; Nath et al., 2012; Azene et al., 2014). LDPE bags or films are usually used in international transportation of fresh fruits (Scheuermann et al., 2014). LDPE is relatively inert and shrinks when heated and is a good barrier for moisture loss while being relatively permeable to O₂, CO₂ and volatiles (Allahvaisi, 2012). The thinner LDPE (25-38 µm) is usually used for shrink-wrapping while the thicker LDPE (45-75 µm) is used for stretch wrapping (Allahvaisi, 2012). HDPE has a higher level of crystallinity due to its non-polar and linear structure and is therefore thicker and stronger compared to LDPE (Allahvaisi, 2012; Bhunia et al., 2013) and therefore acts as a better barrier to the movement of gases and water vapour compared to LDPE (Bhunia et al., 2013). Various researchers have found that HDPE films and bags are effective in reducing moisture loss from fruit during storage (Pongener et al., 2011; Nath et al., 2012; Azene et al., 2014).

5.3. Xtend[®] modified atmosphere/modified humidity (MA/MH)

Xtend[®] films are a specialized type of packaging that have high transmission rates of water vapour (Porat et al., 2009). Compared to poly-ethylene films, Xtend[®] films have higher water vapour transmission rates (Pesis et al., 2000) and is specially designed to eliminate excess moisture that may occur inside the film as a result of condensation (Porat et al., 2009). Because of this, the Xtend[®] film reduces moisture loss and at the same time alleviates problems such as decay which are caused by water condensation inside the packaging. A variety of Xtend[®] films with different transmission rates of O₂, CO₂ and water vapour are available (Porat et al., 2009).

Aharoni et al. (2007) evaluated the effect of Xtend[®] films on nectarine quality during cold storage and found that 'Flamekist' stored in Xtend[®] bags had 0% woolliness whilst over 50% of non-bagged fruit developed woolliness. Porat et al. (2009) also found that Xtend[®] MAP is effective in reducing moisture loss and scald incidence in pomegranates. In addition, Pesis et al. (2000) found that mango fruit packed in Xtend[®] films had low moisture loss and retained their firmness for a longer period compared to fruit packed in micro-perforated polyethylene (PE). Other physiological disorders such as chilling injury, lenticel spot and peel injury were also lower in fruit packed with Xtend[®] films (Pesis et al., 2000). Furthermore, Rodov et al. (2002) found that charentais-type melons packed in Xtend[®] film retained their quality for longer compared to other treatments without Xtend[®] films.

5.4. Edible fruit coatings to reduce moisture loss

An edible coating is a thin layer of edible material that is administered to the surface of a fruit with the purpose of providing an additional barrier against moisture loss (Dhall, 2013). Edible coatings are usually natural polymers obtained from plants and animals and they are mainly made up of proteins, polysaccharides and lipids (Khwaldia et al., 2004; Dhall, 2013). The use of fruit coatings such as wax has been in use for a long time. However, the use of edible coatings is a fairly new technology (Dhall, 2013). Edible coatings can reduce moisture loss from the fruit thus extending the post-harvest life of harvested produce (Khwaldia et al., 2004). In order for edible coatings to be effective in their role of reducing moisture loss, they should be moisture-proof while also allowing sufficient gaseous exchange between the fruit and the environment (Toivonen and Hodges, 2005; Wu, 2010; Becker and Fricke, 2014).

An ideal edible coating is one that is able to reduce moisture loss without negatively affecting the quality of the fruit (Sonti, 2003). Amarante et al. (2001) found that edible coatings offer protection against moisture loss but are less effective in reducing rate of ripening. The effectiveness of an edible coating depends on various factors which include

type of coating, thickness, concentration and pH (Veraverbeke et al., 2003b; Sonti 2003; Dhall 2013). Edible coatings are applied through spraying or dipping (Veraverbeke et al., 2003b) and have the ability to incorporate the flavour of the fruit and therefore they do not alter the taste of the produce (Dhall, 2013). Furthermore, edible coatings are environmentally friendly (Khwaldia et al., 2004; Dhall, 2013). However, some markets remain sceptical on allowing the use of edible fruit coatings.

6. Conclusion

Moisture loss in nectarines is a serious problem that affects the quality of nectarines during storage and shelf life. In addition to fruit mass loss, moisture loss also results in fruit having a shrivelled appearance and this reduces the marketability of the nectarines. Research has been conducted in order to find ways to reduce the moisture loss problem in nectarines but more research is still needed as there are several aspects of post-harvest moisture loss in nectarines that still need to be investigated. These include research on packaging materials that can be used to effectively reduce the moisture loss problem in nectarines. In addition to this, the handling procedures from harvest until consumption needs investigation to come up with ways that can be used to reduce the VPD and minimize moisture loss in the handling chain. Furthermore, the various factors that affect permeability of the nectarine fruit peel still need to be investigated. Such factors include harvest date, tree and orchard effects as well as cultivar differences. Finally the effect of pre-harvest potassium silicate applications also needs to be investigated. By studying moisture loss in a more holistic manner from fruit development until consumption rather than focusing only on packaging, industry will be better equipped to handle and pack nectarines optimally to prevent shrivel incidence.

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PAPER 1:

The effect of fruit to fruit variation, harvest date, tree and orchard effects and cultivar differences on water vapour permeance of nectarine fruit.

ABSTRACT

The research quantified contributions to the total variation in water vapour permeance ($P'H_2O$) from sources such as fruit to fruit differences, cultivar, tree and orchard effects as well as harvest date. The nectarine cultivars used in this study were Alpine, Summer Bright and August Red. The study was carried out during the 2014/2015 and 2015/2016 seasons using five orchards from around the Western Cape Province in South Africa for sampling each cultivar. Fruit were sampled from approx. 4 weeks before the anticipated optimum harvest date until 2 weeks after this date. Overall, fruit to fruit differences were the largest contributor ($> 45\%$) to the total variation in $P'H_2O$ of the three cultivars in both seasons. Harvest date explained less than 35% of the total variation in $P'H_2O$ of the three cultivars. Cultivar effects contributed 7% and 17% to the total variation in the 2014/2015 and 2015/2016 seasons, respectively, while orchard effects contributed 3% to the total variation in both seasons. Tree effects did not contribute anything to the total variation observed in both seasons for the three cultivars overall. Fruit to fruit differences made significant contributions to the total variation in $P'H_2O$ in each of the three individual cultivars in both seasons: $> 48\%$ in 'Alpine', $> 48\%$ in 'Summer Bright' and $> 38\%$ in 'August Red'. Harvest date also made significant contributions to the total variation in $P'H_2O$ in each of the three cultivars in both seasons: $> 34\%$ in 'Alpine', $> 34\%$ in 'Summer Bright' and $> 36\%$ in 'August Red'. Orchard effects only made a small contribution to the total variation in $P'H_2O$: $> 7\%$ for 'Alpine', $> 0\%$ for 'Summer Bright' and 5% for 'August Red'. Tree differences did not contribute to the total variation in $P'H_2O$ in each of the three cultivars. Generally there was an increase in $P'H_2O$ of the fruit peel as the harvest date approached, this means that strategies to minimize moisture loss must be put in place in order to reduce moisture loss and shrivel incidence during storage.

Keywords: Water vapour permeance, moisture loss, shrivel

1. Introduction

Moisture loss is an important post-harvest problem in certain nectarine (*Prunus persica* var. *nectarina*) cultivars. Moisture is lost from the fruit as a result of the vapour pressure deficit (VPD) between the fruit and the surrounding atmosphere. Fruit moisture loss occurs through various parts of the fruit surface, including the stomata, lenticels, cuticle, and epicuticular wax platelets (Díaz-Pérez et al., 2007). Crisosto and Day (2012) found that moisture loss of between 5-8% is sufficient to cause shrivelling in peaches and nectarines during long term storage, rendering them unsaleable (Maguire et al., 2000). Moisture loss can result in huge economic losses as a result of loss in quality and saleable weight (Sastry, 1985). In the 2013/2014 season, South Africa exported about 2,9 million cartons of nectarines with the United Kingdom being the largest export market (54%) followed by Europe and Russia (22%) and the Middle East (19%) (HORTGRO, 2014). The sea freight period to these markets may result in fruit losing moisture and developing a shrivelled appearance.

The fruit peel is the main barrier to moisture loss, but it should also allow sufficient exchange of oxygen and carbon dioxide so that normal metabolic processes continue to take place inside the fruit (Maguire et al., 2001). Maguire et al. (2001) further found that although the fruit peel consist of four layers, the epidermis and the hypodermis are the most permeable layers of the fruit peel. Fruit loses moisture in the form of water vapour which diffuses from the inside to the outside of the fruit as a result of the VPD between the fruit and the surrounding atmosphere (Lara et al., 2014). The ease with which water vapour can escape from a fruit is called the water vapour permeance (P'_{H_2O}) (Maguire et al., 2000). In a constant environment, the P'_{H_2O} of a fruit surface can be calculated from the rate of water loss using Fick's first law of diffusion (Maguire et al., 1999b).

The cuticle is the outermost layer of the fruit peel and prevents excessive loss of moisture from the fruit (Maguire et al., 2001). The efficacy of the cuticle to reduce moisture loss depends on its composition and structure. The composition and structure of the cuticle varies from fruit to fruit depending on the cultivar, harvest maturity, orchard and tree effects, as well as growing conditions (Maguire et al., 2000; Lescourret et al., 2001; Theron, 2015). Maguire et al. (2000) quantified the contribution of each of these factors

to the total variation that is observed in the P'H₂O of apple fruit, while Theron (2015) quantified this in Japanese plums.

Currently no information exists on the P'H₂O of nectarines in South Africa and we therefore decided to determine the P'H₂O of 'Alpine' (susceptible to post storage shrivel), 'Summer Bright' (not susceptible to post storage shrivel) and 'August Red' (highly susceptible to post storage shrivel) nectarines. The research also established if fruit to fruit differences, cultivar differences, harvest maturity, orchard and tree effects have an effect on the P'H₂O of these nectarine cultivars. The main objective was to quantify the contribution of each of these factors to the variation in P'H₂O within a population of harvested nectarine fruit. This information can then be used to develop or re-model post-harvest handling protocols in nectarines so as to minimize post-harvest moisture loss and shrivel incidence in nectarines.

2. Materials and Methods

This trial was divided into two parts. The first part compared peel permeabilities of three nectarine cultivars, namely Alpine (susceptible to postharvest moisture loss), Summer Bright (not susceptible to postharvest moisture loss) and August Red (highly susceptible to postharvest moisture loss), from pre-optimum to post-optimum maturity. The second part investigated how the peel permeability of each of the three cultivars is influenced by various pre-harvest factors.

2.1. Trial sites

The following commercial farms were used for sampling in the 2014/2015 and 2015/2016 seasons: Tandfontein (32°46'18.4"S 19°14'16.5"E), Bokkeveld nursery (33°16'56.1"S 19°19'29.5"E), Verdun (33°16'56.9"S 19°19'29.2"E), Lushof (33°18'02.6"S 19°22'00.7"E), Jagerskraal (33°17'58.4"S 19°19'49.7"E), Timberlea (33°54'24.0"S 18°51'34.9"E), Welgemoed (33°35'01.8"S 18°58'53.4"E), and Nieuwe Sion (33°50'02.5"S 18°57'20.3"E).

2.2. *Experimental layout and sampling*

Five orchards were used for each cultivar and five uniform trees per orchard were randomly chosen. The same orchards were used for sampling in both seasons except for one orchard at Jagerskraal farm (33°17'58.4"S 19°19'49.7"E) which had to be changed due to pest problems. On each sampling date five visually unblemished fruit were harvested from each tree per orchard. Sampling of fruit was done weekly from four weeks before the anticipated optimum harvest date until approximately two weeks after the optimum harvest date. Unfortunately growers sometimes picked all fruit from sample trees during optimum harvest or shortly thereafter, resulting in fewer measurement dates. In order to reduce variation associated with maturity, fruit picked had uniform ground colour. After harvesting, fruit from each tree was carefully placed into a plastic bag and transported with care to the laboratory at the Department of Horticultural Science, Stellenbosch University to avoid injury or disturbance of the fruit peel. The fruit reached the laboratory within 3 to 4 hours after harvest. In the laboratory each fruit was first numbered according to the tree and orchard it was picked from and then the diameter was recorded with a digital calliper (Mitutoyo, Japan). This was done in order to calculate the surface area of the fruit. The shape of the fruit was assumed to be spherical. Afterwards each fruit was weighed using a balance accurate to 0.001 g (XB 320M, Precisa Instruments Ltd., Switzerland). The fruit was then placed in pulp fruit trays and allowed to reach an internal temperature of 20 °C (approx. 5 hours) in a temperature conditioned room. The five fruit per sample were subsequently placed in a plastic container and subjected to an airflow of $\approx 0.5 \text{ m s}^{-1}$ at 20 °C and an average relative humidity (RH) of 60% for a 16 hour period (Fig. 1). Atmospheric RH and temperature was recorded using a Hygrochron™ iButton (CST electronics, Sandton), and pulp temperature was recorded using a Thermocron® iButton. The Hygrochron™ iButton was placed on the underside of the lid of the container to record the RH and air temperature while the Thermocron® iButton was inserted into one fruit which was not part of the experiment, to record pulp temperature at 5 minute intervals during the 16 hour period. Finally, the individual fruit were weighed again after the 16 hour period and the difference in weight was used to calculate the rate of moisture loss, assuming that respiration did not have a significant effect on mass loss due to the relatively short duration over which the test was performed.

2.3. Determination of the water vapour permeance of the fruit peel ($P'H_2O$) of each fruit

The $P'H_2O$ of the fruit peel was calculated using Fick's first law of gas diffusion according to equation 1.

$$P'H_2O = r'H_2O / (\Delta p H_2O * A) \quad (\text{Eq. 1})$$

Where:

$r'H_2O$ = rate of moisture loss (mol s^{-1})

$\Delta p H_2O$ = difference in the water vapour pressure inside and outside the fruit (Pa)

A = area of the fruit surface (m^2)

2.4. Statistical analysis

Contributions of different sources of variation were calculated from mean squares corrected for model effects using components of variance analysis in Dell Inc. (2015) STATISTICA, version 12. The analysis was first done over all three cultivars and then it was done per individual cultivar.

3. Results

3.1. Overall effect of fruit to fruit variation, harvest date, tree and orchard effects and cultivar differences on the peel permeability of all three cultivars

$P'H_2O$ data from both seasons and all three cultivar samples were highly variable. In the 2014/2015 season fruit to fruit differences explained 50% of the total variance obtained overall for all three cultivars (Fig. 2a). In the 2015/2016 season fruit to fruit differences contributed a slightly lower percentage of 45% to the total variance (Fig. 2b). In the 2014/2015 season 40% of the variation was associated with harvest date and in the 2015/2016 season this was 35%. Cultivar effects explained 7% and 17% of the total variance in the 2014/2015 and 2015/2016 seasons, respectively, while orchard effects explained only 3% to the total variance in both the 2014/2015 and 2015/2016 seasons.

Tree effects did not make any contribution to the total variance in both the 2014/2015 and 2015/2016 seasons (Fig 2a and b). In the 2014/2015 season the P'H₂O of 'Summer Bright' increased steadily as the optimum harvest date approached and then slightly decreased after this date (Fig. 3a). 'August Red' displayed a similar steady increase in P'H₂O as the optimum harvest date approached, but then the P'H₂O continued to increase sharply for a week after the optimum harvest date before levelling off in the second week after harvest. For 'Alpine', no sharp increase in the P'H₂O was observed in the 2014/2015 season, the P'H₂O remained almost constant with a slight increase in the P'H₂O observed in the second week after the optimum harvest date. 'August Red' had significantly higher P'H₂O values after optimum harvest date than 'Alpine' and 'Summer Bright' (Fig. 3a). In the 2015/2016 season the P'H₂O of all three cultivars increased until a week before optimum harvest date, but then there was a sharp decrease in the P'H₂O of 'Summer Bright' in the week prior to the optimum harvest date which was followed by a sharp increase in P'H₂O for two weeks after the optimum harvest date (Fig. 3b). 'August Red' displayed a sharper increase in P'H₂O as optimum harvest date approached compared to 'Alpine'. In the 2014/2015 season 'Summer Bright' had a higher P'H₂O (281 nmols⁻¹m⁻²pa⁻¹) at optimum harvest compared to 'Alpine' (160 nmols⁻¹m⁻²pa⁻¹). The P'H₂O of 'August Red' (229 nmols⁻¹m⁻²pa⁻¹) at optimum harvest did not differ significantly from 'Summer Bright' but was significantly higher than 'Alpine' (Fig. 3a). For the 2015/2016 season 'August Red' had the highest P'H₂O on the optimum harvest date (354 nmols⁻¹m⁻²pa⁻¹), followed by 'Alpine' (334 nmols⁻¹m⁻²pa⁻¹) and lastly 'Summer Bright' (288 nmols⁻¹m⁻²pa⁻¹), but these values did not differ significantly from each other (Fig. 3b).

3.2. Effect of fruit to fruit variation, harvest date, tree and orchard effects on the peel permeability of 'Alpine' nectarines

Values of P'H₂O obtained from the entire 'Alpine' sample were highly variable in both the 2014/2015 and 2015/2016 season (Fig. 4). There was a 5-fold difference between the lowest and highest measured P'H₂O in the 2014/2015 season and a 6-fold difference between the lowest and highest measured P'H₂O in the 2015/2016 season (Fig 4). In the 2014/2015, season fruit to fruit differences explained 55% of the total variance while in the 2015/2016 season, fruit to fruit differences explained a slightly lower

percentage (48%) of the total variance (Fig 5a and b). Harvest date explained 34% of the total variance during the 2014/2015 season, but 45% in the 2015/2016 season. Orchard effects explained 11% of the total variance in the 2014/2015 season while in the 2015/2016 season it explained only 7%. In both seasons, tree effects did not contribute to the total variance (Fig 5a and b). Generally, the P'H₂O of 'Alpine' nectarines increased steadily during the sampling period during both seasons (Fig 4a and b). However higher values in P'H₂O were observed in the 2015/2016 season compared to the 2014/2015 season (Fig 4a and b).

3.3. Effect of fruit to fruit variation, harvest date, tree and orchard effects on the peel permeability of 'Summer Bright' nectarines

P'H₂O values obtained for 'Summer Bright' were also highly variable in both seasons (Fig. 6). During the 2014/2015 season, there was a 9-fold difference between the lowest and highest measured P'H₂O whilst in the 2015/2016 season this was a 6-fold difference (Fig 6a and b). Fruit to fruit differences explained 55% of the total variance during the 2014/2015 season while during the 2015/2016 season this was 48% (Fig 7a and b). Harvest date explained 34% of the total variance during the 2014/2015 season and 52% during the 2015/2016 season. Orchard effects explained 11% of the total variance during the 2014/2015 season, but nothing during the 2015/2016 season. Tree differences did not contribute to the total variation of P'H₂O observed in 'Summer Bright' nectarines during both seasons (Fig 7a and b). During the 2014/2015 season, an increasing trend in the P'H₂O of 'Summer Bright' nectarines was observed before the optimum harvest date with a sharp increase in the P'H₂O observed in the week prior to the optimum harvest date. This was observed in all the orchards except for orchard 4 where sample trees were unfortunately harvested by the grower (Fig. 6a). After the optimum harvest date there was a sharp decrease in the P'H₂O from all the remaining orchards for a week after the optimum harvest. However in the 2015/2016 season the trend was different for orchard 2, 3 and 5. In these orchards the P'H₂O increased until one week before optimum harvest and then decreased towards the optimum harvest date. The P'H₂O then continued to increase again steadily after optimum harvest for orchard 2 and 4. For orchard 1 and 3 the P'H₂O increased very slightly for a week after the optimum

harvest date. Sample trees in orchard 5 were unfortunately harvested by the grower and no data is available for the post-optimum fruit.

3.4. Effect of fruit to fruit variation, harvest date, tree and orchard effects on the peel permeability of 'August Red' nectarines

High variability was observed in the $P'H_2O$ for 'August Red' nectarines in both the 2014/2015 and 2015/2016 seasons (Fig. 8). However, unfortunately also for this cultivar, sample trees in some orchards were mistakenly harvested by the growers at the optimum harvest date, orchard 2 in season 1 and in the 2nd season, orchard 3. In the second season, trees from orchard 2 could not be sampled one week after optimum harvest due to a nematicide that was sprayed in the orchard on this particular date. A 5-fold difference was obtained between the lowest and highest measured $P'H_2O$ in the 2014/2015 season whilst a 9-fold difference was obtained in the 2015/2016 season (Fig 8a and b). In the 2014/2015 season 59% of the total variation observed was explained by fruit to fruit differences while only 38% of the total variation was explained by fruit to fruit differences in the 2015/2016 season (Fig 9a and b). 36% of the total variance was explained by harvest date in the 2014/2015 season whilst this was 58% in the 2015/2016 season. Orchard effects explained 5% of the total variance in both the 2014/2015 and 2015/2016 seasons, while tree differences did not make any contribution to the total variance during both seasons (Fig 9a and b). During the 2014/2015 season there was an increasing trend in the $P'H_2O$ during the sampling period from pre- to post-optimum harvested fruit (Fig. 8a). In the 2015/2016 season however the trend was different, with the $P'H_2O$ increasing steadily towards the optimum harvest date and then decreasing after the optimum harvest date (Fig. 8b).

4. Discussion

Overall, for all three nectarine cultivars fruit to fruit differences made the biggest contribution (45% and 50% for two seasons) to the total variation in $P'H_2O$ observed during both seasons. Theron (2015) also found that fruit to fruit variation was the biggest contributor (> 45%) to the total variation in $P'H_2O$ of 'African Delight™', 'Laetitia' and

'Songold' plums. However, Maguire et al. (2000) found that harvest date rather than fruit to fruit differences was the biggest contributor to the total variation in P'H₂O of 'Braeburn' fruit. Fruit to fruit differences also explained most of the variation observed in the individual cultivars (> 50% in Alpine, > 55% in Summer Bright and > 59% in August Red). Fruit to fruit differences may be due to factors such as differences in maturity of fruit sampled on the same date, position of fruit within the canopy, fruit shape and size as well as fruit contact with shoots or leaves during growth (Maguire et al., 1999a; Theron, 2015). Maguire et al. (1999a) found that position of apple fruit within the canopy did influence the P'H₂O with fruit from more exposed areas of the canopy having higher P'H₂O values compared to fruit from inner canopy areas. In this study fruit sampling did not take place on dedicated bearing positions since the fruit was sampled based on ground colour and size only. Fruit shape and size may also have an effect on the P'H₂O because they affect the distribution of stress on the fruit surface and this in turn affects both the degree of micro-cracking on the fruit surface and the fracture patterns (Gibert et al., 2007). Micro-cracks increase the P'H₂O of fruit and they also act as entry sites for pathogens (Gibert et al., 2007). Fruit contact with shoots or leaves can result in damage of the fruit peel and cuticle resulting in increased P'H₂O of the fruit. Theron (2015) proposed that stricter methods in choosing which fruit to harvest may not be the solution to reduce the large fruit to fruit differences since commercial harvesting is done by hand with fruit being picked and sorted based on visual appearance (ground colour and size). This still results in large fruit to fruit variation within cartons leading to a large variability in the incidence of shrivel. Since the fruit size and ground colour might not give a correct indication of maturity at harvest, industry should consider using non-destructive measures to sort fruit in order to obtain more even fruit maturities in the carton.

Harvesting date made the second largest contribution (> 35%) to the total variance in P'H₂O for all three cultivars. Fruit maturity was not determined on each sampling date since fruit sampling started four weeks before the anticipated commercial harvest date, at this stage the fruit was obviously immature. It is also important to note that the uniformity in maturity amongst harvested fruit was important since the goal was to harvest fruit of similar maturities on each sampling date. Therefore, determining fruit maturity on a sample of fruit on each sampling date was not going to be an accurate indicator of

similar maturity levels amongst harvested fruit. In both seasons it was observed that there was generally an increase in P'H₂O in 'Alpines', 'Summer Bright' and 'August Red' nectarines with later sampling dates. The same observation was also made in individual analysis of the three cultivars. Harvest date explained greater than 34% of the total variance obtained in both seasons for 'Alpine' nectarines. For 'Summer Bright' and 'August Red' nectarines, harvest date was the biggest contributor to the total variation in P'H₂O in the 2015/2016 season contributing greater than 52% and greater than 58%, respectively. This is important to note since the result suggests that these cultivars must be handled carefully after harvest to prevent excessive moisture loss. Although 'Summer Bright' is not prone to shrivel, the fruit quickly loses its fresh, glossy appearance and assume a dull skin colour due to moisture loss (personal observation). 'August Red' is highly prone to shrivel and any losses in moisture can cause significant reduction in quality of the fruit. The increase in P'H₂O as fruit matured beyond their harvest date was also observed by Maguire et al. (2000) in apple and Theron (2015) in Japanese plums. Sastry (1985) postulated that over-mature fruit will lose moisture more rapidly than mature fruit due to tissue aging. This was the case in our trial since the average P'H₂O of all three cultivars generally continued to increase after the optimum harvest date. Maguire et al. (2000) found a similar effect in 'Braeburn', 'Pacific Rose', 'Cripps Pink', and 'Granny Smith' apples and concluded that the increase in P'H₂O with later harvest dates may be due to a number of inherent growth and environmental factors. Although there was a general increasing trend in the P'H₂O as we moved towards the harvesting date, there were some orchards which did not follow this trend. In the 2014/2015 season 'Summer Bright' displayed a very sharp increase in P'H₂O in the week prior to the optimum harvest date and this was followed by a sharp decrease in P'H₂O in the week after optimum harvest date. The reason for this is not clear and further research is needed in this regard. The increase in P'H₂O as the fruit matures can also be explained by the presence of micro-cracks which become more pronounced as the fruit matures (Peschel and Knoche, 2005; Gibert et al., 2007). As the fruit remain on the tree after optimum maturity, fruit tissues, including the cuticle waxes, begin to break down and wax crystals become degraded due to effects of wind, sun and mechanical abrasion, this causes damage to

the barriers for moisture loss leading to increased $P'H_2O$ in over-mature fruit (Maguire et al., 2001).

Cultivar differences explained more than 7% of the total variation in $P'H_2O$ of the three nectarine cultivars. Variation in $P'H_2O$ amongst different cultivars may be caused by variation in the occurrence of micro-cracks and differences in soluble cuticular lipids and their molecular structure (Lescourret et al., 2001). In nectarines the development and occurrence of micro-cracks changes as the fruit develops with most micro-cracks occurring during the final growth stage (Gibert et al., 2007). In addition to this, the micro-cracks that occur during the early stages of peach growth do not close and continue to spread over the fruit surface during fruit growth and development (Lescourret et al., 2001). Cline et al. (1995) also found that surface conductance varied amongst sweet cherry cultivars as a result of variation in the occurrence of micro-cracks. In addition to differences in the occurrence of micro-cracks amongst different cultivars, variation in $P'H_2O$ amongst cultivars is also related to differences in the physical and chemical properties of outer layers of the fruit, cuticle thickness as well as amount and type of epicuticular waxes (Maguire et al., 2000). 'August Red' is the most susceptible to shrivel followed by 'Alpine', while 'Summer Bright' is the least prone to shrivel. If $P'H_2O$ were the only factor that influenced shrivel one would expect a clear correlation between highest $P'H_2O$ and shrivel incidence. In the 2014/2015 season however, 'Summer Bright', although not susceptible to shrivel had the highest average $P'H_2O$ while 'Alpine' had the lowest average $P'H_2O$ with 'August Red' in between. Therefore the reason for the higher incidence in shrivel of 'Alpine' should be found elsewhere. The reason could be related to fruit size, 'Alpine' is generally far smaller in fruit size (about 56 mm in diameter at optimum harvest) as it is an early season cultivar and hence has a higher surface area to volume ratio and is more sensitive to moisture loss.

Orchard effects explained only 3% of the total variation in $P'H_2O$ for both seasons. This shows that the orchards were relatively uniform and therefore did not make a big contribution to the variation in $P'H_2O$. Although the differences amongst the orchards were small, these differences may have been due to different orchard management practices. According to Crisosto et al. (1994) 'O'Henry' peaches from excessively irrigated

orchards lost 30% more moisture compared to fruit from optimally irrigated orchards. The reason for this was explained by Gibert et al. (2007) who found that the development of micro-cracks varied amongst orchards depending on the level of irrigation, with high levels of irrigation on 'Zephyr' nectarines increasing density of micro-cracks on fruit compared to water-restricted trees. The high density of micro-cracks should lead to high $P'H_2O$ values in fruit from excessively irrigated orchards. Orchard effects did also not contribute much to the total variation in $P'H_2O$ observed in the individual cultivars (> 7% in 'Alpine', 11% in 'Summer Bright' and 5% in 'August Red'. As found by Maguire et al. (2000) for apples and Theron (2015) for plums, tree differences did not contribute to the total variation in $P'H_2O$. The reason for this might be that $P'H_2O$ is a fruit characteristic and influences from the whole tree physiology are very small (Maguire et al., 2000).

5. Conclusion

The study showed that the peel of 'Summer Bright' nectarine is generally more permeable to water vapour compared to 'August Red' and 'Alpine', indicating that $P'H_2O$ does not fully explain shrivelling since 'Summer Bright' is the least shrivel prone amongst the three cultivars. The study also showed that the $P'H_2O$ of the fruit increased as the harvest date approached. With this in mind, it is important to implement measures which help to reduce moisture loss at harvest. Such measures include harvesting fruit in the cooler time of the day, keeping fruit under a shade after harvesting and covering fruit with wet blankets. To further minimize moisture loss and shrivel, fruit should be cooled as soon as possible after harvest to remove field heat. Although the study showed that less mature fruit have lower $P'H_2O$ values, it cannot be recommended to harvest less mature fruit since other factors such as attainment of acceptable eating qualities and susceptibility to internal disorders during cold-storage should also be considered. However, fruit should be harvested as soon as they reach their optimum maturity and unnecessary delays in harvesting should be avoided. The study also showed that large fruit to fruit variation was the main contributor to the variation in $P'H_2O$. The large fruit to fruit variations make it difficult to effectively manage the fruit so as to reduce moisture loss and shrivel incidence. However, there is a need for further research to establish the extent to which other factors

such as position of fruit within canopy, fruit contact with shoots and leaves, exposure to sunlight, size and shape of the fruit influence the fruit P'H₂O. It is therefore important to implement measures which can help to obtain fruit of more similar maturities within a carton. Such measures include the use of non-destructive methods to determine fruit maturity. There is also need for further research to establish the critical moisture loss point at which different cultivars will begin to shrivel since the P'H₂O of individual fruit varies depending on the cultivar.

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7. Figures

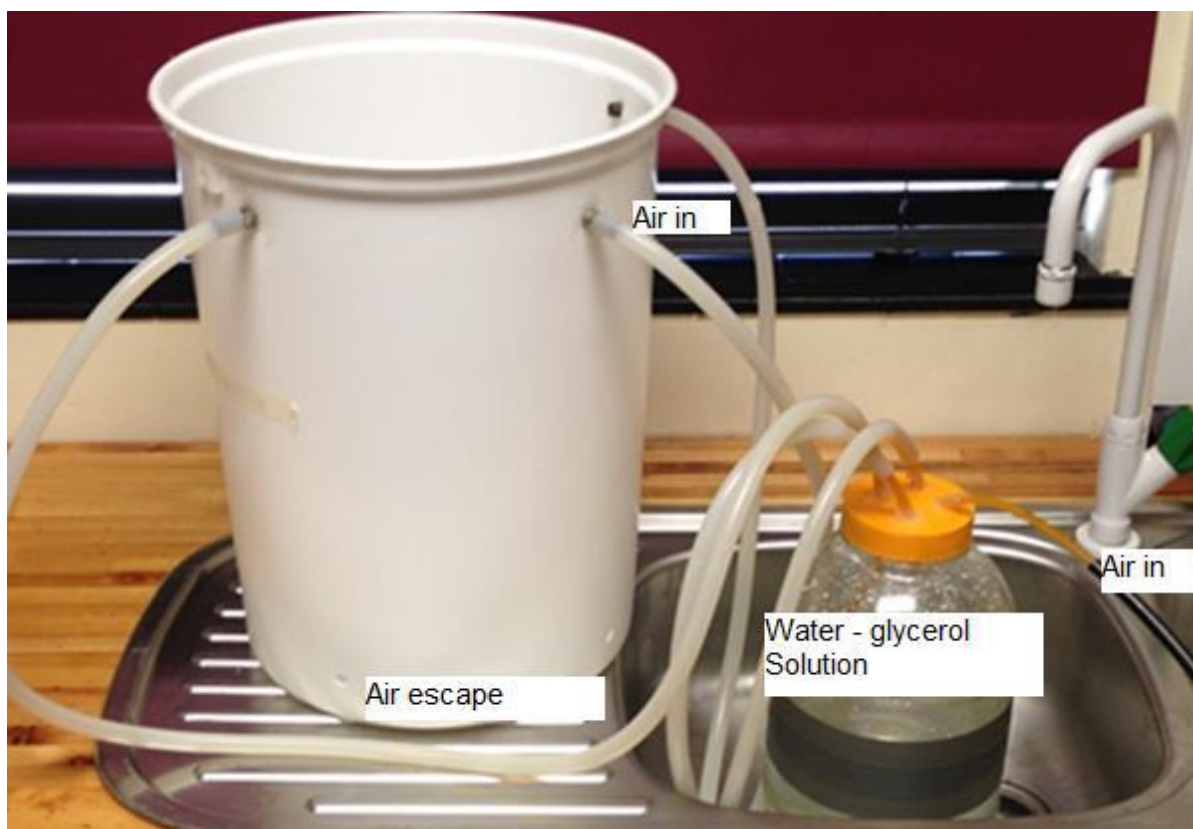


Fig. 1. An illustration of the system used in this research to determine the P'H₂O of nectarine fruit peels. Air from a compressor was bubbled through a glycerol and water solution to adjust the RH of the air to 60%. The air was subsequently forced at $\sim 0.5 \text{ m s}^{-1}$ over the fruit in the container and escaped through the holes in the bottom of the container (With permission from Theron, 2015).

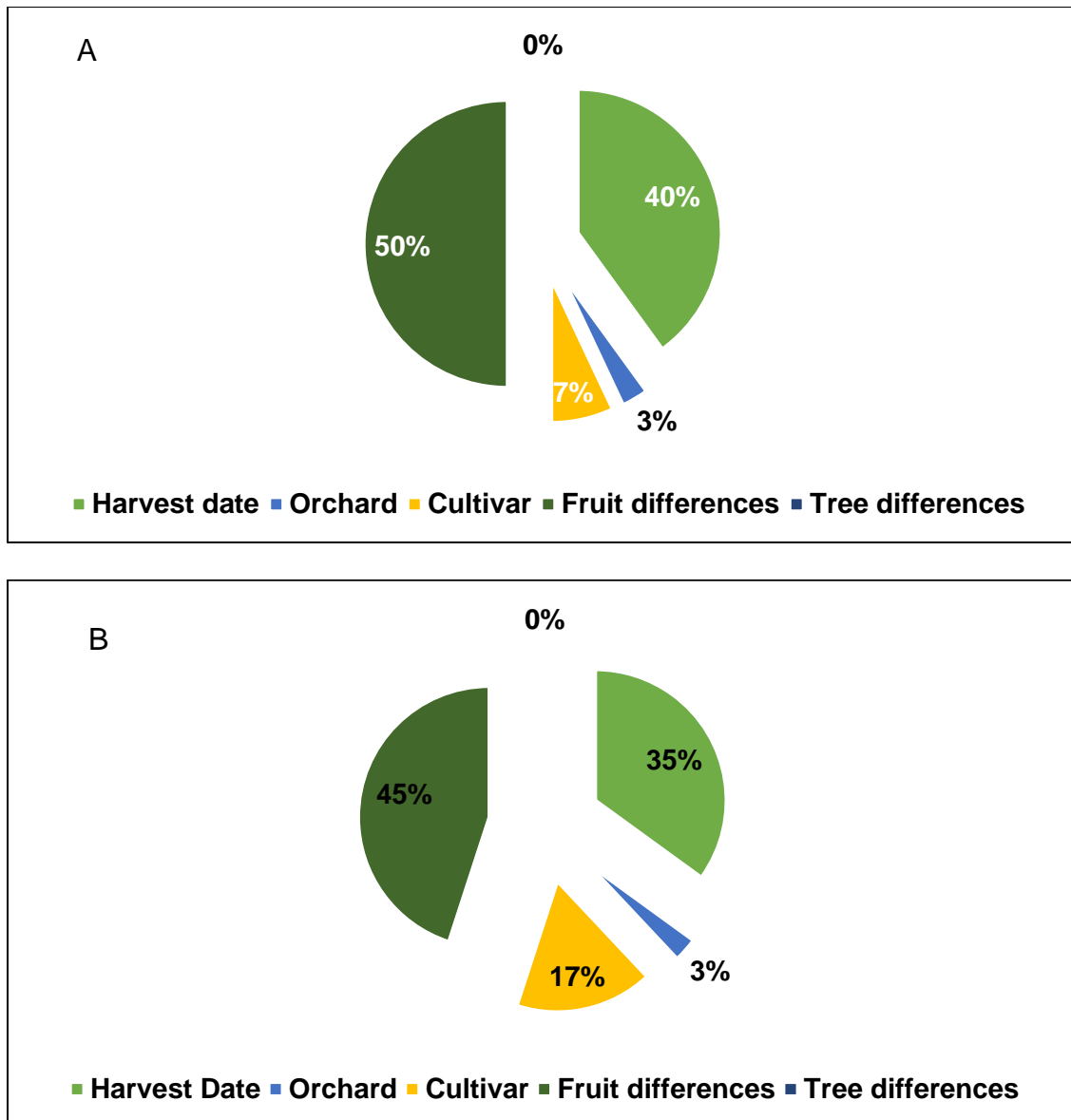


Fig. 2. Relative components of variance as contributed by fruit to fruit variation, orchard differences, harvest dates, cultivar and tree to tree differences to the total variation of the water vapour permeance of three nectarine cultivars ('Alpine', 'Summer Bright' and 'August Red'). The fruit were sampled from 4 weeks prior to the optimum harvest date until approximately 2 weeks after the optimum harvest date from commercial farms. Fig. 2a represents the 2014/2015 season and Fig. 2b represents the 2015/2016 season.

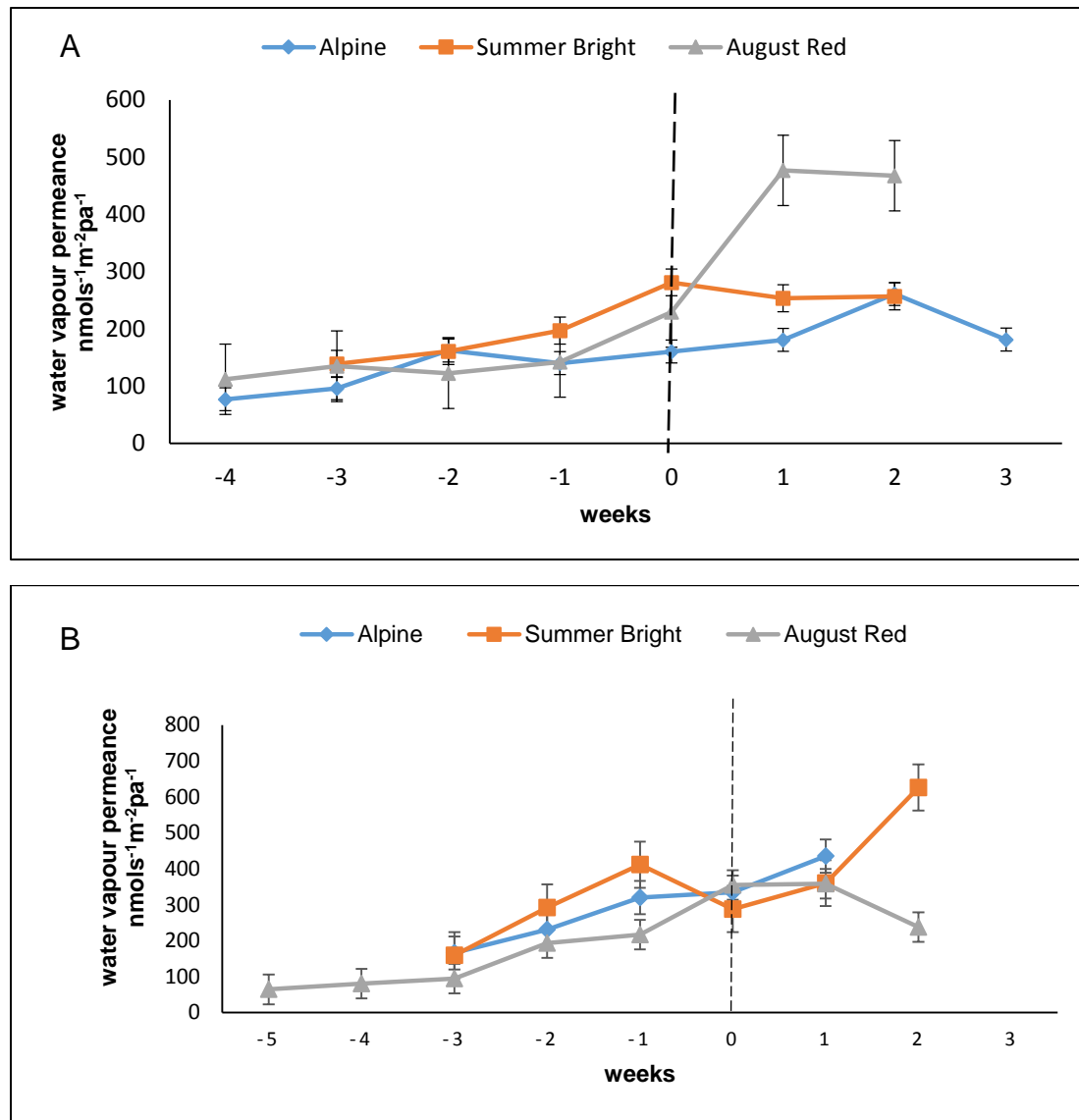


Fig. 3. Water vapour permeance of ‘Alpine’, ‘Summer Bright’ and August Red’ from 4 weeks before harvest date to 3 weeks after the optimum harvest date. The data presented is the mean of five orchards’ data per cultivar. Fig. 3a represents the 2014/2015 season and Fig. 3b represents the 2015/2016 season.

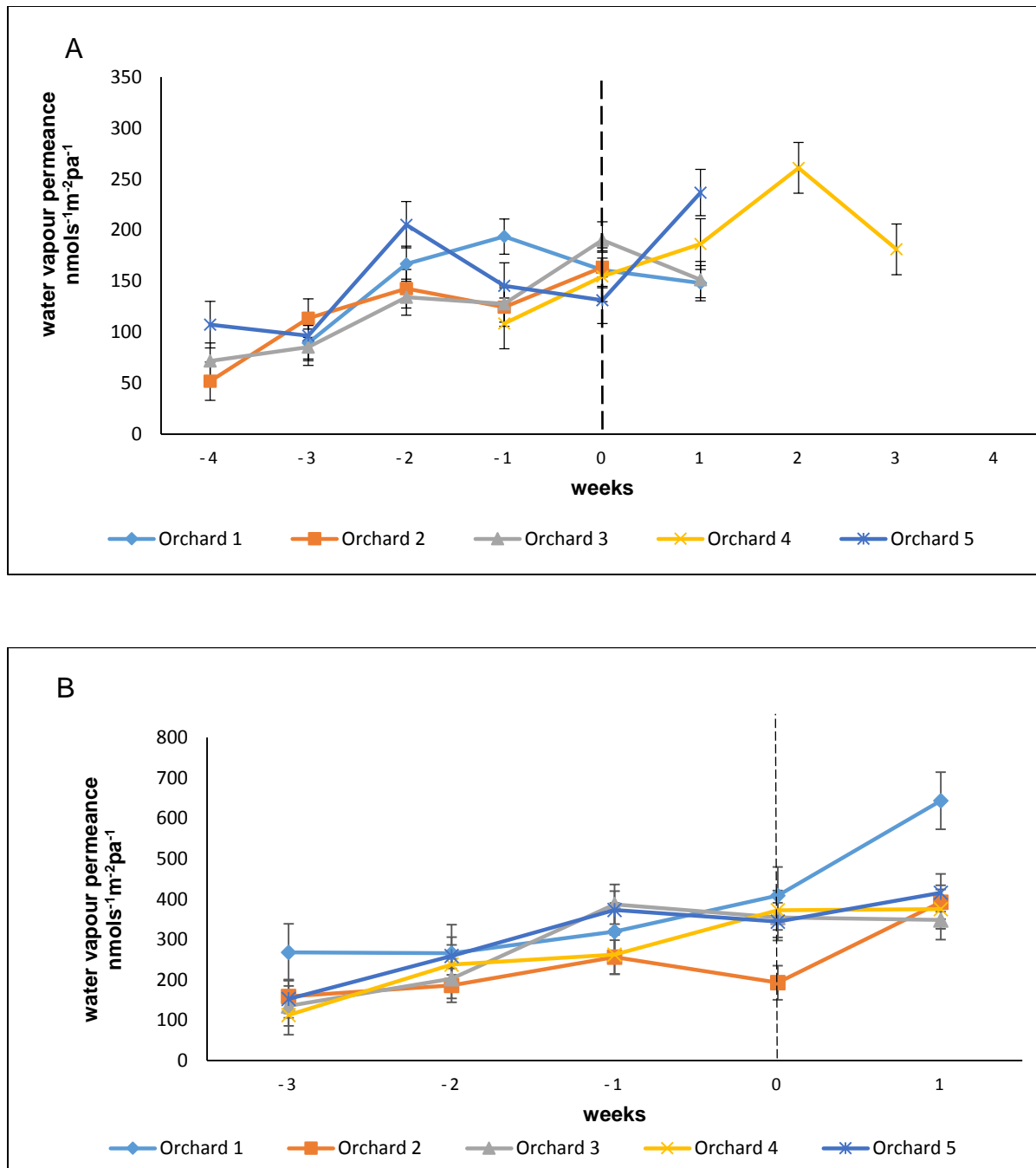


Fig. 4. Water vapour permeance of 'Alpine' fruit from five orchards harvested at different times relative to the commercial harvest date. Fig. 4a represents the 2014/2015 season and Fig. 4b represents the 2015/2016 season.

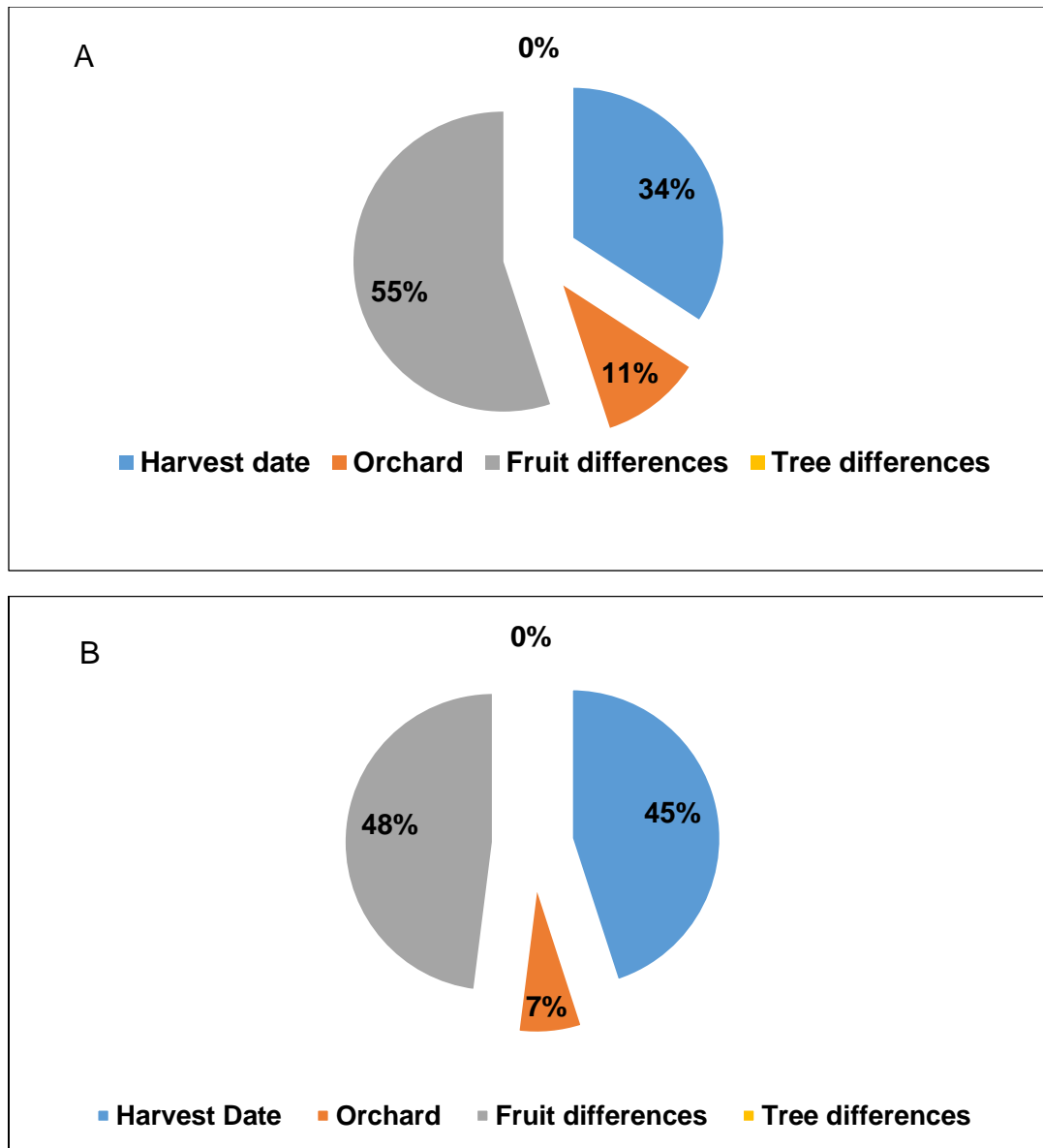


Fig. 5. Relative components of variance as contributed by fruit to fruit variation, orchard differences, harvest dates and tree to tree differences to the total variation of the water vapour permeance of 'Alpine' nectarines. The fruit were sampled from five commercial orchards during from 4 weeks prior to the optimum harvest date until approximately 2 weeks after the optimum harvest date from five commercial orchards. Fig. 5a represents the 2014/2015 season and Fig. 5b represents the 2015/2016 season.

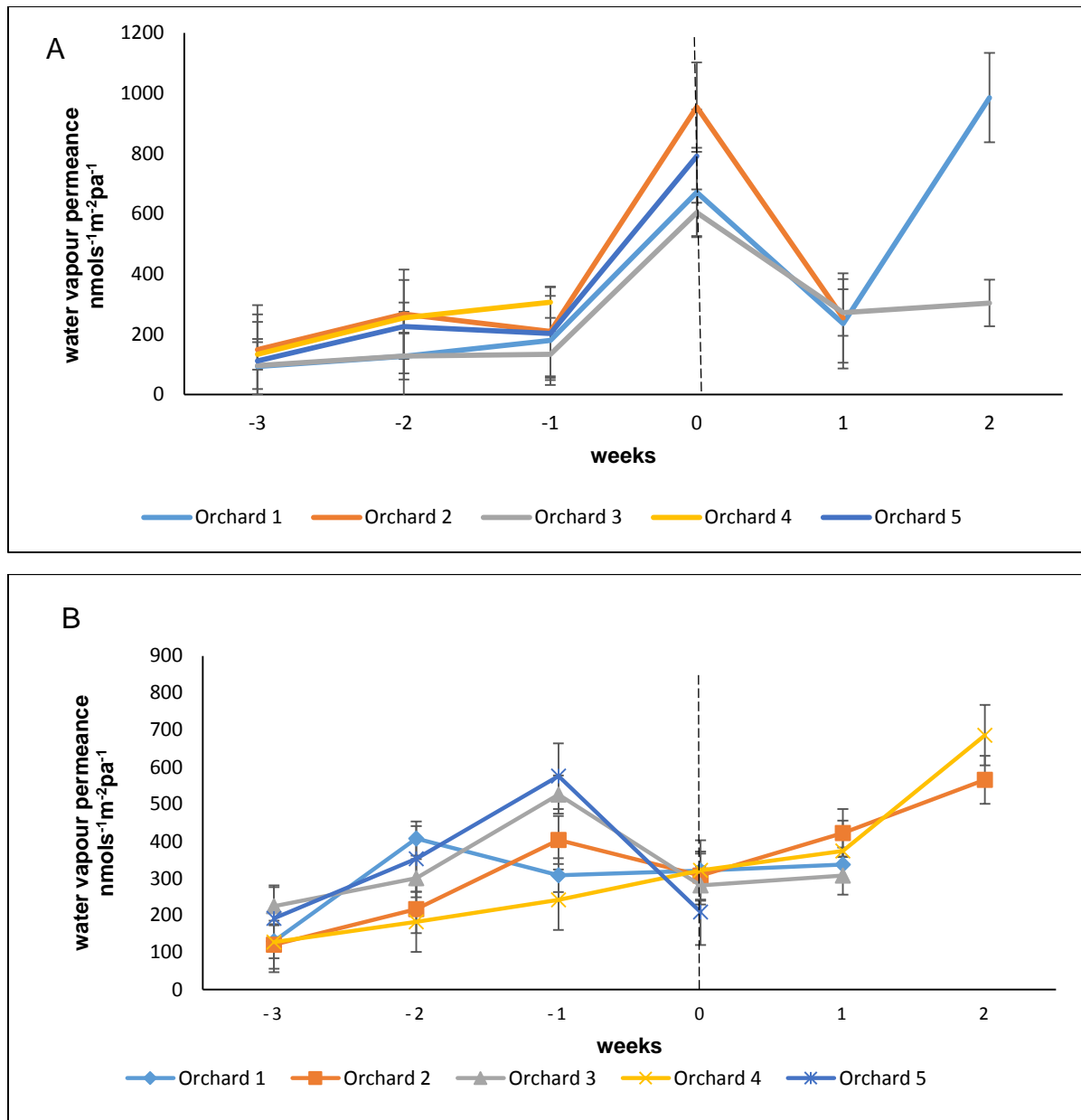


Fig. 6. Water vapour permeance of 'Summer Bright' fruit from five orchards harvested at different times relative to the commercial harvest date. Fig. 6a represents the 2014/2015 season and Fig. 6b represents the 2015/2016 season.

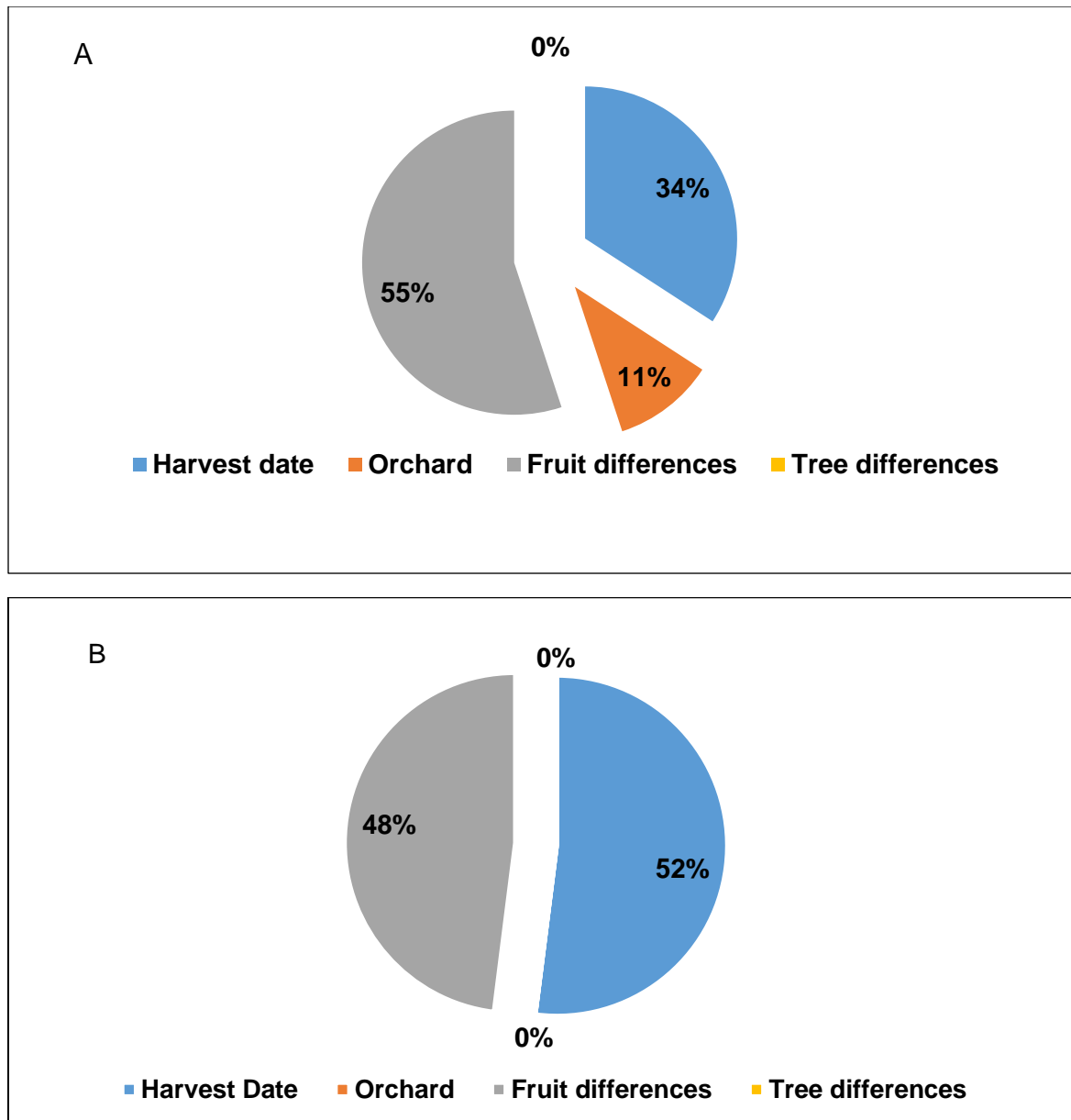


Fig. 7. Relative components of variance as contributed by fruit to fruit variation, orchard differences, harvest dates and tree to tree differences to the total variation of the water vapour permeance of 'Summer Bright' nectarine cultivar. The fruit were sampled from five commercial orchards from 4 weeks prior to the optimum harvest date until approximately 2 weeks after the optimum harvest date. Fig. 7a represents the 2014/2015 season and Fig. 7b represents the 2015/2016 season.

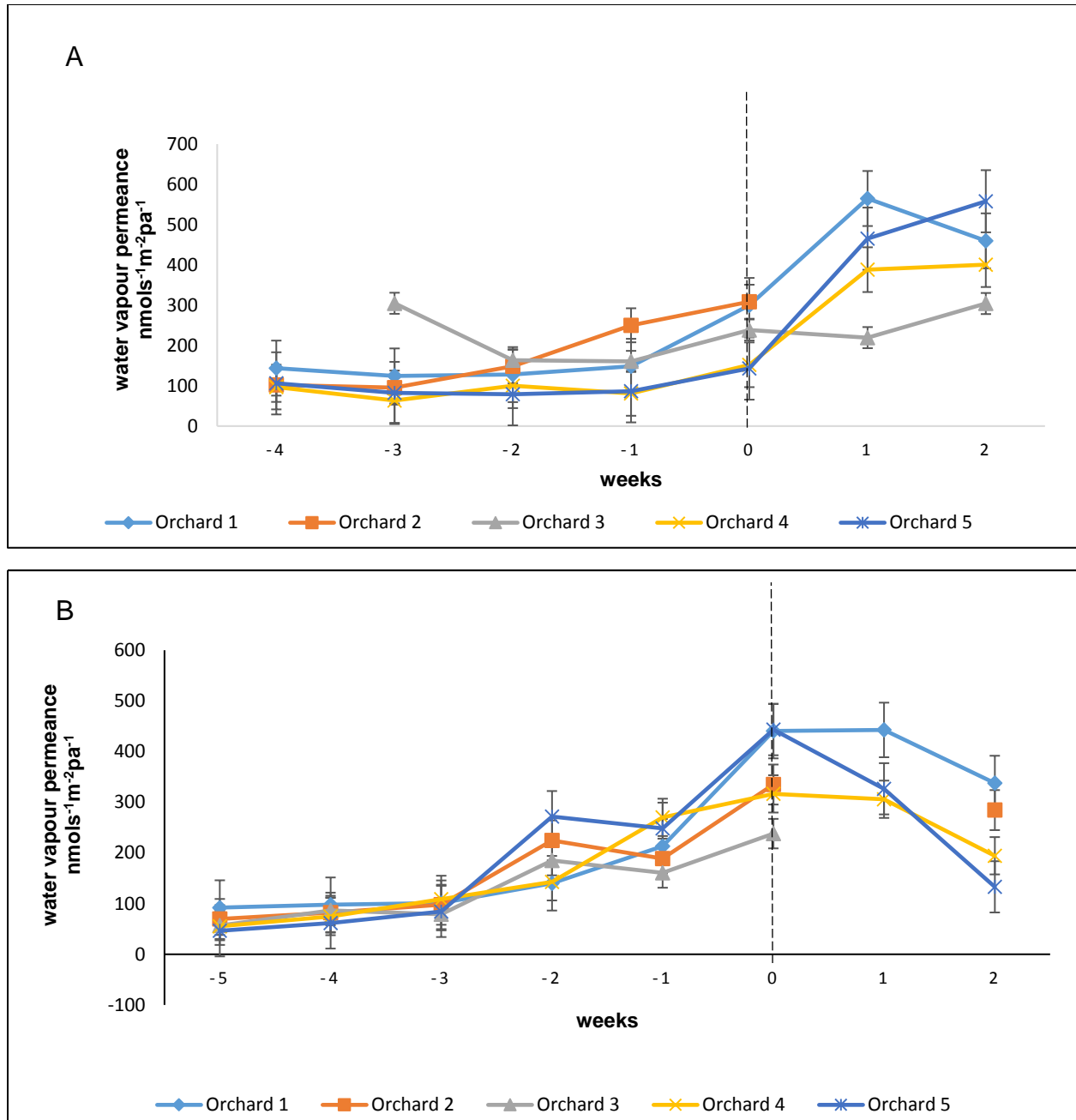


Fig. 8. Water vapour permeance of 'August Red' fruit from five orchards harvested at different times relative to the commercial harvest date. Fig. 8a represents the 2014/2015 season and Fig. 8b represents the 2015/2016 season.

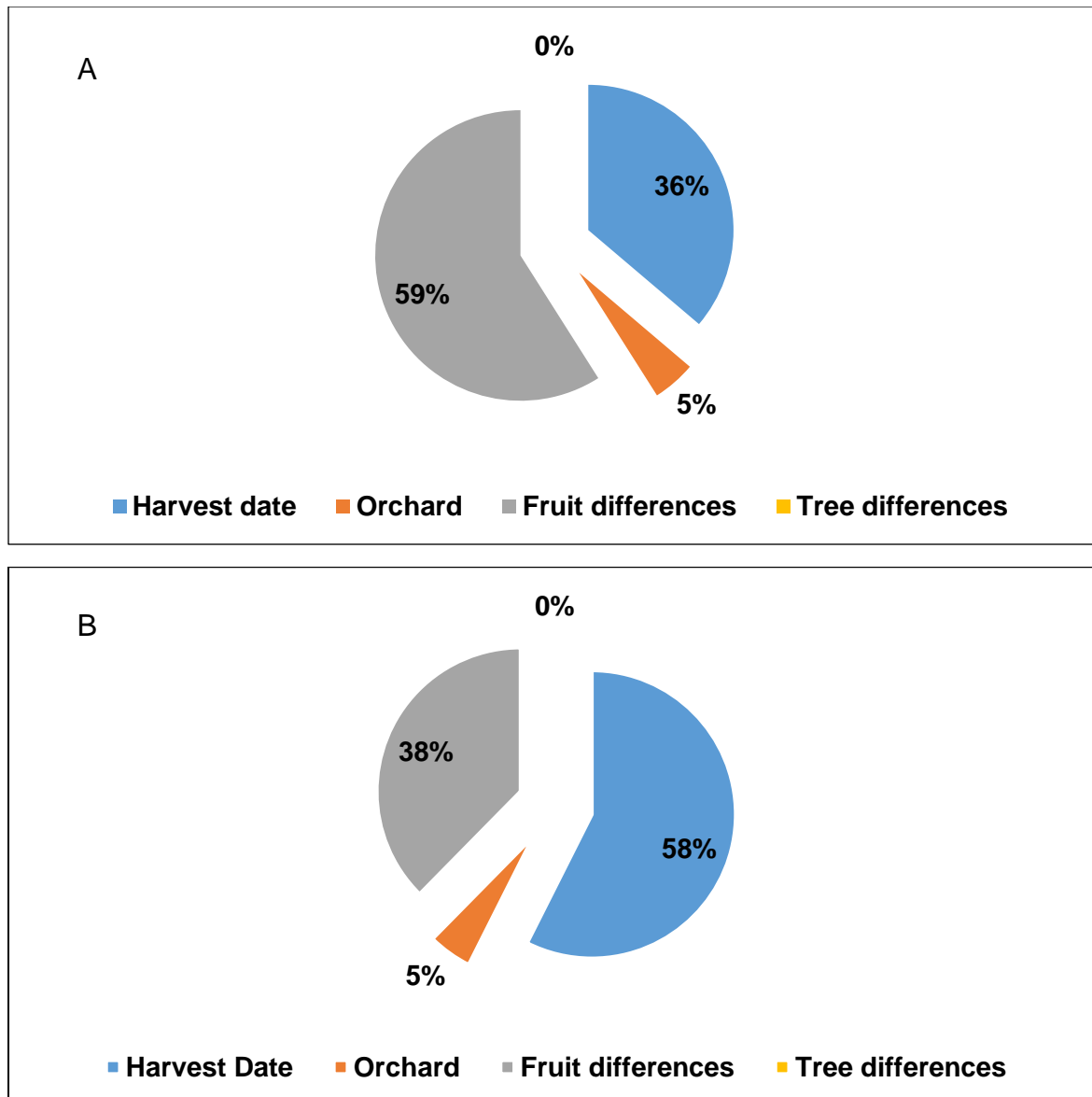


Fig. 9. Relative components of variance as contributed by fruit to fruit variation, orchard differences, harvest dates and tree to tree differences to the total variation of the water vapour permeance of 'August Red' nectarines. The fruit were sampled from five commercial orchards from 4 weeks prior to the optimum harvest date until approximately 2 weeks after the optimum harvest date. Fig. 9a represents the 2014/2015 season and Fig. 9b represents the 2015/2016 season.

PAPER 2:

The contribution of vapour pressure deficit to post-harvest mass loss and post-storage shrivel manifestation in ‘August Red’ nectarines (*Prunus persica* var. *nectarina*) in different handling protocols

ABSTRACT

The South African stone fruit industry is export oriented and fruit are subjected to long handling chains from harvest until they eventually reach their markets. This exposes fruit to moisture loss conditions as a result of the vapour pressure deficit (VPD) between the fruit and surrounding atmosphere. Moisture loss results in loss of saleable weight as well as fruit having a shrivelled appearance when they reach the market. The aim of this study was to determine the VPD for different handling chains from harvest until the end of shelf-life and to establish where in the handling chain the risk for moisture loss is the highest and whether it can be mitigated by changes to the protocols. The study was carried out in the 2014/2015 and 2015/2016 growing seasons on ‘August Red’ nectarines, a shrivel sensitive cultivar, and consisted of five different handling chains, viz., the control (recommended industry handling protocol), pre-ripening treatment in which fruit was pre-ripened before forced-air cooling (FAC) and 3 other treatments in which fruit was pre-cooled at 0 °C for different durations i.e. 24 hours, 48 hours and 72 hours. Fruit quality was evaluated at harvest, after cold-storage and after shelf-life. Mass loss was determined on arrival at the pack house, after forced air cooling (FAC), after cold-storage and after shelf-life. The VPD was significantly higher in the pre-ripening treatment in both seasons and this resulted in fruit from this treatment losing more moisture although not significantly more than from the fruit which was pre-cooled for 24 hours and 72 hours, respectively. The total mass loss from harvest until end of shelf-life was significantly lower in the control and fruit pre-cooled for 48 hours. The high temperature (20 °C) to which fruit were exposed to during pre-ripening resulted in fruit losing significantly more moisture during the period between arrival at the pack house and the end of FAC. However, this did not translate to a higher incidence of shrivel after both cold-storage and cold storage followed by shelf-life. Pulpiness and woolliness were significantly higher in the pre-ripening treatment after cold storage followed by shelf life compared to the other

treatments. It is therefore recommended not to pre-ripen August Red nectarines but rather to rapidly cool fruit after harvest in order to prevent pulpiness and woolliness. This study indicated that the currently recommended industry handling protocols should be carefully followed in order to minimise the VPD and hence mass loss throughout the handling chain.

Keywords: Vapour pressure deficit, moisture loss, mass loss, handling chain, shrivel

1. Introduction

The South African stone fruit industry is mainly oriented towards export and in the 2013/2014 season approximately 2.9 million cartons of nectarines were exported with the United Kingdom being the largest export market (54%), followed by Europe and Russia (22%) and the Middle East (19%) (HORTGRO, 2014a). Nectarine fruit usually spend four weeks in cold storage during loading, accumulation of containers and shipment to overseas markets and have a shelf-life of 5-7 days (Laubscher, 2006). The South African fruit export handling chains consists of many steps and role-players from harvest until fruit reaches the consumers (Goedhals-Gerber et al., 2015). These long handling chains expose nectarines to post-harvest moisture loss conditions and may affect the quality of nectarines when they finally reach their markets. At harvest, fruit have a fresh appearance and crisp texture. However, harvesting removes the fruit from its water supply and the fruit will begin to lose moisture with this moisture not being replaced (Goedhals-Gerber et al., 2015; Holcroft, 2015). Moisture loss after harvesting produce has an immediate economic impact in that it leads to loss of saleable weight and can also result in shrivel incidence (Holcroft, 2015; Sastry, 1985).

The fruit peel is the main barrier to moisture loss, but it should also allow sufficient exchange of oxygen and carbon dioxide so that normal metabolic processes continue to take place inside the fruit (Maguire et al., 2001). Fruit moisture loss occurs through various parts of the fruit surface, these include the stomata, lenticels, cuticle and epicuticular wax platelets (Díaz-Pérez et al., 2007). Fruit loses moisture in the form of water vapour which diffuses from the inside to the outside of the fruit as a result of the vapour pressure deficit

(VPD) between the fruit and the surrounding atmosphere (Lara et al., 2014). VPD describes the driving force for moisture loss from the produce to the surrounding atmosphere and it is increased by increasing temperature while decreasing with increasing relative humidity (RH) (Holcroft, 2015). In a constant environment, the rate of water loss from produce can be calculated using Fick's first law of diffusion (Maguire et al., 1999).

In order to effectively reduce moisture loss from harvested produce, it is important that the VPD between fruit and the surrounding atmosphere is kept to a minimum at each stage of the handling chain (Holcroft, 2015). It is also important to reduce the time between harvesting and pre-cooling so as to reduce the duration when the fruit is exposed to a high VPD. In addition to reducing moisture loss from fruit, pre-cooling of fruit also helps to reduce the respiration rate of fruit and this ensures a longer shelf-life of the fruit (Paull, 1999).

Currently the handling protocol for nectarines in South Africa is to harvest fruit during the cooler time of the day (temperature under 25 °C) and to remove field heat as soon as possible after harvesting by cooling fruit to just above the dew point temperature of the pack-house (HORTGRO, 2014b). Furthermore, the handling protocol requires that nectarines should be packed within 12 hours of arrival at the pack-house (HORTGRO, 2014b). Low temperature disorders such as woolliness and pulpiness (Internal breakdown) limit the cold-storage life of nectarines and reduce the quality of nectarines on arrival in the markets (Lurie and Crisosto, 2005). Pre-ripening of nectarines has been done for over 50 years and involves a delay in the commencement of cooling by keeping fruit at 20 °C for approximately 48 hours after harvest (Laubscher, 2006; Nanos and Mitchell, 1991). The use of pre-ripening to reduce incidence of pulpiness and woolliness has been reported by several researchers (Laubscher, 2006; Lurie and Crisosto, 2005; Nanos and Mitchell, 1991) and in this study we added a pre-ripening stage to one of the treatments to see if pre-ripening can indeed reduce internal breakdown without increasing shriveling in 'August Red' nectarines.

The aim of this study was to determine the VPD between 'August Red' nectarines and their environment during different simulated post-harvest handling chains. This

information will show where in the handling chain the risk for moisture loss is the highest, and with this information at hand the industry will be better equipped to create and apply optimum handling protocols to prevent moisture loss.

2. Materials and methods

2.1. Fruit sampling and experimental layout

The trial was conducted in the 2014/2015 and 2015/2016 seasons on 'August Red' nectarines. The fruit were sampled from Tandfontein farm (32°46'18.4"S 19°14'16.5"E), Koue Bokkeveld, Western Cape, South Africa. Five treatments were used to simulate different handling chains from harvest to packing (Table 1) and each treatment was replicated 6 times. On the commercial harvest date, visually unblemished fruit of the same size and ground colour were harvested and placed into lug boxes. Eighteen lug boxes were used per treatment (3 lugs per replicate) with approximately 25 fruit harvested into each lug box. The weight of the fruit of two lugs per replicate was determined as soon as possible after harvest in the orchard as well as on arrival at the laboratory. The other lug box per replicate was used to determine fruit maturity in the laboratory. A Thermocron® iButton (CST electronics, Sandton) recording fruit pulp temperature was inserted into one fruit (an extra fruit that was not used for any other measurements) per replicate (lug) at harvest. A Hygrochron™ iButton, which recorded air temperature and RH, was placed on the inside of each lug. The iButtons were numbered according to their treatment and replicate, accompanied the fruit in their respective replicates throughout the different simulated handling chains to facilitate the calculation of the VPD of each treatment and replicate. Temperature and RH was recorded every 15 min in the 2014/15 season and every hour in the 2015/16 season from harvest until the end of shelf-life. After harvesting fruit in the field was completed, the lug boxes were transported to the laboratory at ExperiCo (PO Box 4022, Idas Valley, Stellenbosch, 7609, South Africa). The transport duration was approximately 2.5 h in an uncooled vehicle to simulate transport of fruit from the orchard to the pack-house. After the different delay periods depicted in Table 1 (pre-ripening or cooling at 0 °C), fruit were packed according to export standards in 2.5 kg

single layer traypack interlock cartons and lined with a perforated (54 x 2 mm) high density polyethylene (HDPE) bag with a thickness of 16 μm before the commencement of the 24 h forced air cooling (FAC) period. Therefore for each treatment, two cartons were packed, one for cold storage and one for cold storage + shelf-life. The weight of the fruit from each of the two cartons per treatment was again determined after FAC. After the 4-week cold storage period at $-0.5\text{ }^{\circ}\text{C}$ the HDPE bags were removed from the cartons and the fruit were placed in simulated shelf-life conditions for 5 days at $10\text{ }^{\circ}\text{C}$.

2.2. Fruit evaluation

On the commercial harvest date, 25 fruit from one lug box per replicate per treatment were used to determine the maturity of the fruit. Hue angle was determined on both cheeks of 5 fruit per replicate using a calibrated colorimeter (Minolta colour recorder DR-10, Japan). Flesh firmness (kg) was determined on both peeled cheeks of 10 fruit per replicate using a FTA (Fruit Texture analyser, Güss Instruments) fitted with an 11 mm tip. Total soluble solids (TSS, %Brix) was determined on a pooled juice sample of 25 fruit per replicate using a temperature controlled, digital refractometer (Palette, PR-32 ATAGO, Bellevue, USA). Total titratable acidity (TA, %) was determined on a pooled juice sample of 25 fruit per replicate. TA was determined by titrating a 10 g aliquot of the juice sample with 0.1 M NaOH to a pH end-point of 8.2 using an automated titrator (Metrohm AG 760, Herisau, Switzerland).

The weight of fruit from each of the two cartons was determined after the respective cold storage and shelf-life periods. Fruit quality was also determined after cold-storage and after cold-storage + shelf life. After cold-storage, all packaging films were removed from one carton per replicate per treatment, this is the carton that was destined for the simulated shelf-life period of 5 days at $10\text{ }^{\circ}\text{C}$. All the fruit from the other carton was inspected for the incidence of shrivel (%) and decay (%). A fruit was considered to be shrivelled when the shrivelled skin extended over the shoulder of the fruit. Hue angle and flesh firmness was determined on 5 fruit as described above. Internal defects were determined by cutting the rest of the fruit per replicate around the equatorial axis, separating the two halves of the fruit. If the fruit pulp had a dry texture with no free juice

when the fruit halve was squeezed, the fruit was classified as woolly. However, if the fruit had a dry texture with a little free juice running from the pulp when the fruit halve was squeezed, the fruit was classified as pulpy. If a fruit had internal or pit cavity browning it was classified as having internal browning. Total chilling injury (CI), was seen as the sum of the woolly, pulpy and fruit with internal browning per replicate. Fruit was classified as overripe (OR) when abnormally soft with excessive amounts of free juice and when the mesocarp tissue in the sub-epidermal region developed a translucent breakdown while the inner tissue exhibited a normal appearance. Total internal defects was calculated by adding the total CI and OR.

2.3. Statistical analysis

Data were analysed with a mixed model repeated measures analysis of variance using Dell Inc. (2015) STATISTICA. ANOVA generated P-values and the significant differences between means were determined using Fisher's least significant differences (LSD) test with a 95% confidence interval.

3. Results

Maturity indexing carried out in both seasons showed that the treatments generally did not differ significantly from each other in terms of the quality and maturity parameters measured at harvest (Table 2 and 3). However, TA was significantly lower in the 72h at 0 °C treatment in the 2014/2015 season, while the other four treatments did not differ significantly from each other (Table 2).

3.1. Mass loss

The mass loss data for the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on mass loss. Treatments significantly interacted with evaluation times in percentage mass loss at different time intervals (Fig. 1). Mass loss did not differ significantly during the first period (between harvest and arrival at the pack house) for the different treatments as all

treatments were handled the same during this period. The pre-ripened treatment lost significantly more mass (3.8%) compared to all the other treatments in the period between arrival at the pack house and the end of FAC (Time 2). During this same period, the mass loss in the 48h at 0 °C treatment did not differ significantly from that in the 72h at 0 °C treatment, but was significantly higher compared to the mass loss in the control and the 24h at 0 °C treatments. During the cold-storage period (Time 3), mass loss was significantly more in the 24h at 0 °C treatment (2.8%) than in any other treatment. During this same period, the mass loss in the control, pre-ripening and 72h at 0 °C treatments were not significantly different from each other, but were significantly higher compared to the mass loss in the 48h at 0 °C treatment. Therefore, the 48h at 0 °C treatment resulted in the least mass loss during the cold storage period. During the shelf-life period (Time 4), the 72h at 0 °C treatment significantly lost more mass (1.9%) compared to the other four treatments. However, that mass loss in the other four treatments during this same period were not significantly different from each other.

For the control treatment, the mass lost during Time 1 and Time 2 were not significantly different. However, significantly more mass was lost during Time 3. The mass lost during Time 4 for the control treatment was significantly lower than that of Time 3 while it was significantly higher than that of Time 1 (Fig. 1). For the pre-ripening treatment Time 2 had a significantly higher mass loss and this was followed by Time 3 which did not differ from Time 4 and lastly Time 1 which had the lowest mass loss. For the 24h at 0 °C treatment, Time 3 resulted in a significantly higher mass loss compared to the other three time intervals, which did not differ from each other in mass loss. For the 48h at 0 °C treatment, Time 2, 3 and 4 lost more mass and did not significantly differ from each other while Time 1 resulted in the lowest mass loss. For the 72h at 0 °C treatment, Time 2, 3 and 4 also lost more mass and were not significantly different from each other whilst being significantly different from Time 1 which had the lowest mass loss.

Treatments also significantly interacted with evaluation times in accumulated mass loss at specific times during the handling chains (Fig. 2). On arrival at the pack-house, there were no significant differences in the mass lost amongst all treatments. Significant differences in the mass loss only emerged after FAC, when the pre-ripening treatment

had a higher accumulated mass loss (5.4 g/fruit) at the end of FAC, but this was not significantly different from the 72h at 0 °C treatment. The mass lost in the control, 24h at 0 °C, 48h at 0 °C were not significantly different from each other at the end of FAC. By the end of cold-storage, the pre-ripening treatment still had the highest accumulated mass loss, but was not significantly different from the 24h at 0 °C and 72h at 0 °C treatments. The 48h at 0 °C treatment had the lowest mass loss (5.5 g/fruit) at the end of cold storage and was not significantly different from the mass loss in the control treatment at this stage. At the end of shelf-life, the pre-ripening treatment still had the highest accumulated mass loss (9.4 g/fruit), but was not significantly different from the 24h at 0 °C and 72h at 0 °C treatments. The control and the 48h at 0 °C treatments had significantly lower mass loss at the end of shelf-life.

When the total mass loss from harvest until the end of shelf-life was expressed as a percentage of the initial mass, the pre-ripening, 24h at 0 °C and 72h at 0 °C treatments lost most mass, but did not differ from each other. The control and the 48h at 0 °C treatments lost less mass and did not differ from each other (Fig. 3). However, the total mass lost in the 24h at 0 °C and in the 48h at 0 °C treatments were also not significantly different.

3.2. Vapour Pressure Deficit (VPD)

The total VPD varied significantly amongst the treatments in both the 2014/2015 and 2015/2016 seasons (Fig. 4 and Fig. 5, respectively). However, the data loggers were not able to record data for the whole duration from harvest until the end of shelf life during the 2014/2015 season. This was because the data loggers quickly reached their maximum recording capacity due to a smaller recording interval (15 minutes) that was used. Because of this, a longer recording interval of 1 hour was used in the 2015/2016 season and the loggers were able to capture data for the whole duration. During the 2014/2015 season, the pre-ripening treatment had a significantly higher total VPD (878 mbar), this was followed by the control, 48h 0 °C and 72h 0 °C treatments with the 24h 0 °C treatment having the lowest total VPD (405 mbar) (Fig. 4). During the 2015/2016 season the pre-ripening treatment again had the highest total VPD (524 mbar), but was

not significantly higher than the control treatment (321 mbar) whilst being significantly different from the other three treatments (Fig. 5).

3.3. *Quality Parameters*

In both seasons fruit quality parameters were measured at harvest, after cold-storage and after shelf-life. The data obtained after cold storage and shelf life from the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit quality. No incidence of decay, soft tips, pulpiness, woolliness and internal browning were found after cold storage. The incidence of shrivel and overripeness after cold-storage were low and did not differ significantly among the treatments (Table 4). After cold storage hue angle did not differ significantly among the treatments. However, flesh firmness differed significantly with the pre-ripening treatment having a significantly lower firmness (9.8 kg) compared to the other four treatments which did not differ significantly from each other.

The evaluations after cold-storage + simulated shelf life did not show any incidence of decay, shrivel, soft tips and internal browning. The hue angle of the control, pre-ripening and 24h at 0 °C treatments were highest and did not differ significantly from each other (Table 5). The hue angle for the pre-ripening treatment did not differ significantly from that of the 48h at 0 °C and 72h at 0 °C treatments. Flesh firmness of the pre-ripening treatment remained significantly lower (6.30 kg) compared to the other four treatments which did not differ significantly from each other. Pulpiness and woolliness (21.4% and 14.7%, respectively) was significantly higher in the pre-ripened fruit compared to the other four treatments which did not significantly differ from each other. It is possible that the low woolliness levels in treatments 1, 3, 4 and 5 could be ascribed to the fruit still being relatively firm and woolliness only occurs in nectarines after ripening. Although overripeness was recorded after cold-storage + simulated shelf life, it did not differ significantly among the different treatments.

4. Discussion

In the period between harvest and arrival at the pack-house (Time 1), fruit from all treatments were handled in the same manner and no significant differences in percentage mass loss were seen among the treatments during this period. The small, non-significant differences that were recorded may be due to fruit to fruit differences in peel permeability as highlighted by Theron (2015) and also in Paper 2 of this study. These fruit to fruit differences in peel permeability will cause fruit to lose moisture at variable rates even though exposed to similar conditions. In the period between arrival at the pack-house and the end of FAC, the pre-ripening treatment lost more mass compared to the other four treatments. The reasons for this high mass loss in the pre-ripening treatment would be due to the fruit being subjected to a relatively high pre-ripening temperature of 20 °C for approximately 48 hours during pre-ripening and that the fruit were not pre-cooled to remove field heat. Elevated temperatures increase the water VPD which is one of the driving forces for moisture loss between the fruit and the storage atmosphere, and this will subsequently lead to increased mass loss (Paull, 1999; Theron, 2015). A direct correlation between mass loss and VPD was also reported by Whitelock et al. (1994). In both seasons the VPD between the fruit and the storage atmosphere was significantly higher in the pre-ripening treatment. The high mass loss in the pre-ripening treatment indicates that the 85 % RH in the pre-ripening room as well as the high density perforated shrivel sheets in which the fruit was packed were inadequate to prevent excessive moisture loss from the fruit. According to Crisosto (2005), if pre-ripening of fruit is not properly monitored, it may result in excessive moisture loss, excessive softening and shrivelling of stone fruit. Due to the high percentage mass loss during Time 2, the pre-ripening treatment had a higher accumulated mass loss at the end of FAC.

During Time 2, treatment 3 in which the fruit were kept at 0 °C for 24 hours after arrival at the pack-house, had the lowest mass loss and was not significantly different from the control treatment. The beneficial effect of pre-cooling fruit before packaging were also observed by Wijewardane and Guleria (2013) in apples, where pre-cooling reduced moisture loss in apples if a high RH was maintained in the cold room. Pre-cooling of produce helps to remove field heat from fruit and this reduces the respiration rate as well

as the VPD between the fruit and the surrounding atmosphere subsequently reducing mass loss from the produce (Maguire et al., 2001; Martínez-Romero et al., 2003). Although pre-cooling fruit for 24 hours before packaging resulted in lower mass loss during Time 2, it should also be noted that pre-cooling fruit for 48 hours and 72 hours did not further reduce mass loss but actually resulted in higher mass loss. This shows that pre-cooling of 'August Red' nectarines may be beneficial when fruit is kept at 0 °C for only 24 hours. Theron (2015) also found a general increase in mass loss in Japanese plums as the pre-cooling time increased in fruit kept at 0 °C. The reason for this observation is not clear but it may be due to the low RH at 0 °C. Although the pre-ripening treatment lost a significantly higher amount of moisture during Time 2, the mass lost during cold storage (Time 3) in this treatment was significantly lower. The mass lost from the pre-ripening treatment during cold storage was not significantly different from mass lost in the control and 72 h at 0 °C treatments. This shows that the pre-ripening conditions were responsible for the high mass loss in Time 2 for the pre-ripening treatment. The percentage mass loss during the shelf life period (Time 4) was not significantly different for all the treatments except for the 72 h at 0 °C treatment which had a significantly higher mass loss during this period. For all the treatments, the percentage mass loss during shelf life (Time 4) was the lowest in comparison to mass loss in the other 3 time periods, this may be because the fruit had already lost a lot of moisture during the earlier stages in the handling chain and only a small amount of moisture could still be lost during shelf-life.

Although the main aim of the study was to determine the effect of mass loss on shrivel manifestation in 'August Red' nectarines, very little shrivel incidence was recorded after cold storage and no shrivel incidence was recorded after cold storage + shelf-life. The reason for this may be that the amount of moisture lost by the nectarines was not high enough to cause shrivel incidence. According to Holcroft (2015) peaches and nectarines have a water content of about 89% and will only show symptoms of shrivel when they have lost at least 19% of this water. In our study the nectarines only lost an average mass of 5% and this was therefore not sufficient to induce shrivel. Evaluations carried out after cold-storage did not show any incidence of decay, soft tips, pulpiness, woolliness and internal browning. Although shrivel and overripeness were recorded after cold-storage, these did not differ significantly among the treatments. The flesh firmness

of the pre-ripening treatment was significantly lower compared to the other four treatments after both cold storage and cold storage + simulated shelf-life. The reason for this is that the pre-ripening induces cell wall disassembly which continues slowly throughout storage so that at the end of cold storage the pre-ripened fruit will be softer compared to fruit which was not pre-ripened (Infante et al., 2009). Although the aim of pre-ripening was to soften fruit to ± 6.5 kg, this was difficult due to high fruit to fruit variation. This resulted in fruit having average firmness values higher than 6.5 kg even after cold storage. Laubscher (2006) also found that fruit from the same cultivar may differ in their response to pre-ripening conditions.

Even though no shrivel incidence was recorded after cold storage + simulated shelf-life, internal breakdown (IB) particularly pulpiness and woolliness were recorded after this period. The pre-ripened fruit had a significantly higher percentage of pulpiness and woolliness after cold storage + simulated shelf life compared to the other four treatments. IB of fruit became visible during shelf-life because IB normally appears when fruit begin to ripen after a long period of cold storage (Zoffoli et al., 2002). Crisosto et al. (1995) found that fruit that lose water more readily are at a higher risk of developing chilling injury symptoms such as IB. This was the case in our study since the pre-ripened treatment had a higher VPD in both seasons and a higher total mass loss percentage. Crisosto et al. (2004) and Infante et al. (2009) showed that pre-ripening of peach cultivars before cold storage has the capacity to reduce the incidence of IB, which was not the case in our study as the pre-ripened fruit had a higher incidence of IB. A possible reason for this observation is that the fruit may have been harvested relatively immature because the average flesh firmness in both seasons was above the acceptable maximum for 'August Red' nectarines which is 11.3 kg (DAFF, 1998). Immature fruit will not ripen properly leading to a higher incidence of physiological disorders such as IB (Crisosto et al., 1995; Tijskens et al., 2007). In addition to this, the positive effect of pre-ripening in reducing pulpiness and woolliness is cultivar specific and these results show that 'August Red' cultivar does not respond well to pre-ripening. The development of pulpiness and woolliness in nectarines during shelf-life has been attributed to changes in the activity of pectin methylesterase (PME) as well as reduction in the activity of endo-polygalacturonase (endo-PG) during cold-storage (Lurie and Crisosto, 2005).

5. Conclusion

The total mass loss from harvest until end of shelf-life was lowest in the control and 48h at 0 °C treatments while the other three treatments had a higher total mass loss percentage and did not differ significantly from each other. The pre-ripening treatment lost significantly more mass in the period between arrival at the pack-house and the end of FAC. The reason for this is that the fruit was exposed to a pre-ripening temperature of 20 °C for approximately 48 hours and this increased the VPD between the fruit and the surrounding atmosphere causing fruit to lose more moisture. In both seasons the total VPD was significantly higher for the pre-ripening treatment. It is important to limit the time during which fruit is exposed to this high pre-ripening temperature so that moisture loss is kept to a minimum. During this same period, the control and fruit which was kept at for 24h at 0 °C after arrival at the pack-house had a significantly lower moisture loss. However, it was also found that keeping fruit at 0 °C for 48 hours and 72 hours after arrival at pack-house did not further reduce mass loss but actually increased mass loss. This shows that extended periods of pre-cooling, for longer than 24 hours may not be beneficial in reducing mass loss in 'August Red' nectarines.

There was no significant incidence of shrivel after both cold-storage and cold storage + shelf-life, this is because the amount of moisture lost was not high enough to induce shrivel in 'August Red' nectarines. However, IB particularly pulpiness and woolliness were recorded after cold storage + shelf-life and were significantly higher in the pre-ripening treatment. The effectiveness of pre-ripening in reducing IB is cultivar specific and this study showed that 'August Red' does not respond well to pre-ripening. It is therefore recommended not to pre-ripen August Red nectarines and to rapidly cool fruit after harvest in order to prevent further deterioration of fruit and avoid incidence of IB. Other methods to prevent woolliness, such as controlled atmosphere storage needs to be investigated.

From the results of this study it was clear that none of the proposed handling chains performed better than the control (recommended industry handling protocol) in terms of reducing moisture loss, shrivel and internal breakdown. It is therefore recommended that

the industry handling protocol should be carefully followed so that moisture loss and its associated problems are kept to a minimum.

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7. Tables and Figures

Table 1.

Treatments used to simulate different handling chains of nectarines in South Africa.

Treatment	Harvest	Transport to pack-house	Period at 0 °C before packing	Pack into commercial packaging	Pre-ripening ¹	Forced air cooling ²	Cold storage ³	Shelf-life ⁴
1	✓	✓		✓		✓	✓	✓
2	✓	✓		✓	✓	✓	✓	✓
3	✓	✓	24 h	✓		✓	✓	✓
4	✓	✓	48 h	✓		✓	✓	✓
5	✓	✓	72 h	✓		✓	✓	✓

¹Pre-ripen to ± 6.5 kg at 20 °C and 85 % relative humidity

²Forced air cooling for 24 h

³Cold storage at -0.5 °C for 4 weeks

⁴Simulated shelf-life period of 5 days at 10 °C

Table 2.

Quality parameters of 'August Red' nectarines at harvest (2014/2015 season). Explanation of the treatments is presented in Table 1.

Treatment	Hue angle	Flesh firmness (kg)	Total Soluble solids	Titratable Acidity
Control	49.1 ns	12.7 ns	15.1 ns	1.10 a
Pre-ripening	48.9	12.6	15.1	1.11 a
24h 0 °C	47.0	12.7	15.4	1.08 a
48h 0 °C	48.6	12.7	15.0	1.08 a
72h 0 °C	47.6	12.5	14.5	1.01 b
<i>P-value</i>	<i>0.422</i>	<i>0.742</i>	<i>0.111</i>	<i>0.045</i>
<i>LSD (P≤0.05)</i>	<i>2.593</i>	<i>0.393</i>	<i>0.692</i>	<i>0.069</i>

Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test.

Table 3.

Quality parameters of 'August Red' nectarines at harvest (2015/2016 season). Explanation of the treatments is presented in Table 1.

Treatment	Hue angle	Flesh firmness (kg)	Total soluble solids	Titratable Acidity
Control	47.43 ns	11.9 ns	14.00 ns	1.02 ns
Pre-ripening	48.98	11.6	13.38	0.98
24h 0 °C	47.19	11.5	13.43	1.00
48h 0 °C	47.09	11.6	13.27	1.01
72h 0 °C	43.66	11.3	13.17	0.98
<i>P-value</i>	<i>0.494</i>	<i>0.493</i>	<i>0.167</i>	<i>0.719</i>
<i>LSD (P≤0.05)</i>	<i>6.387</i>	<i>0.562</i>	<i>0.711</i>	<i>0.080</i>

Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test.

Table 4.

Quality of 'August Red' nectarines after cold storage of 4 weeks at -0.5 °C. The data of the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit quality. Explanation of the treatments is presented in Table 1.

Treatment	Hue angle	Flesh firmness (kg)	Shrivel (%)	Overripe (%)
Control	51.91 ns	12.5 a	0.00 ns	0.00 ns
Pre-ripening	50.68	9.8 b	0.00	0.00
24h 0 °C	49.45	12.2 a	0.46	0.00
48h 0 °C	48.29	12.5 a	0.00	0.46
72h 0 °C	49.85	11.5 a	0.93	0.00
<i>P-value</i>	<i>0.494</i>	<i>0.000</i>	<i>0.240</i>	<i>0.416</i>
<i>LSD (P≤0.05)</i>	<i>4.911</i>	<i>1.116</i>	<i>0.985</i>	<i>0.587</i>

Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test.

Table 5.

Quality of 'August Red' nectarines after cold-storage of 4 weeks at -0.5 °C plus a simulated shelf-life period of 5 days at 10 °C. The data of the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit quality. Explanation of the treatments is presented in Table 1.

Treatment	Hue angle	Flesh firmness (kg)	Pulpy (%)	Woolly (%)	Overripe (%)
Control	49.62 a	10.8 a	4.17 b	2.38 b	0.98 ns
Pre-ripening	47.59 ab	6.3 b	21.39 a	14.67 a	0.00
24h 0 °C	50.46 a	10.7 a	2.30 b	1.19 b	0.00
48h 0 °C	43.27 b	10.3 a	4.24 b	0.60 b	0.00
72h 0 °C	43.67 b	10.5 a	4.66 b	1.79 b	0.49
<i>P-value</i>	<i>0.037</i>	<i>0.000</i>	<i>0.000</i>	<i>0.000</i>	<i>0.240</i>
<i>LSD (P≤0.05)</i>	<i>5.668</i>	<i>1.664</i>	<i>7.398</i>	<i>6.461</i>	<i>1.043</i>

Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test.

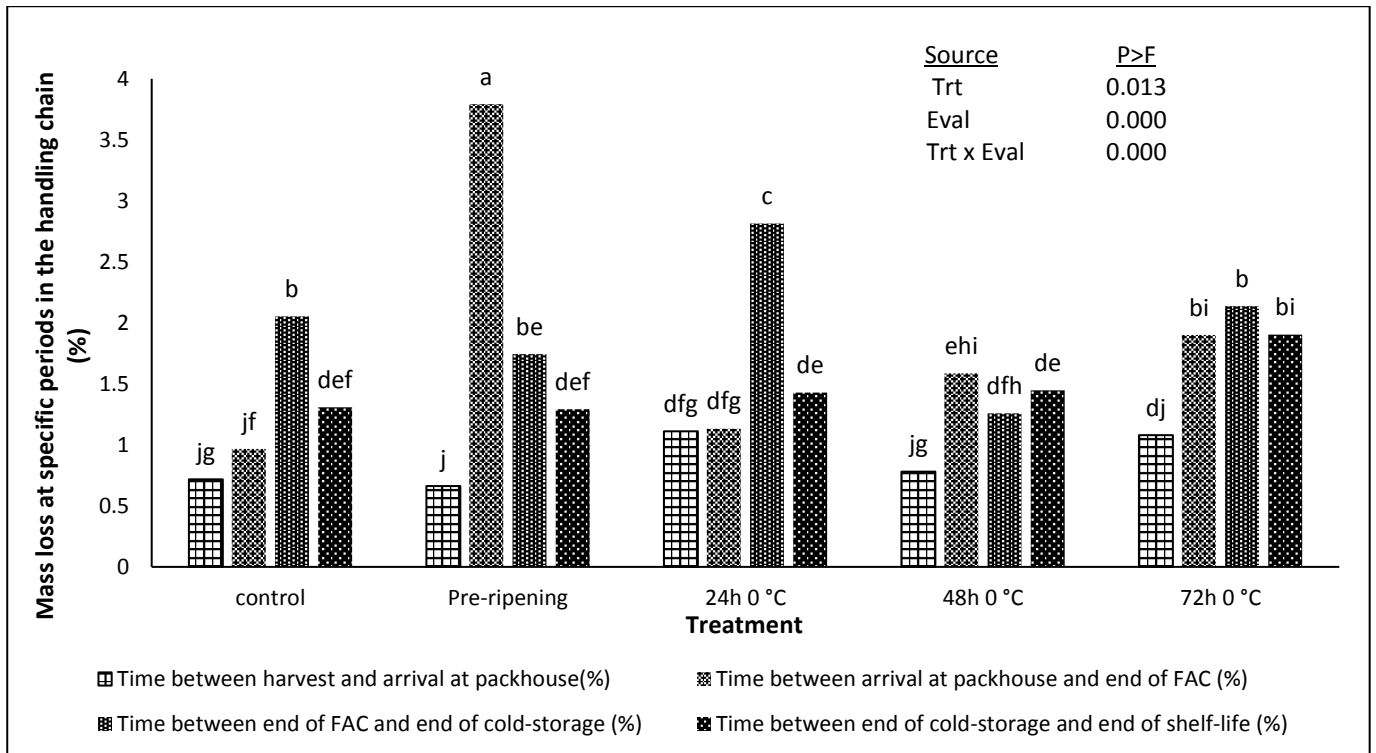


Fig. 1. Percentage mass loss at specific periods in the handling chain for 'August Red' nectarines. The data of the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit mass loss. Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test. Explanation of the treatments is presented in Table 1. FAC = forced air cooling.

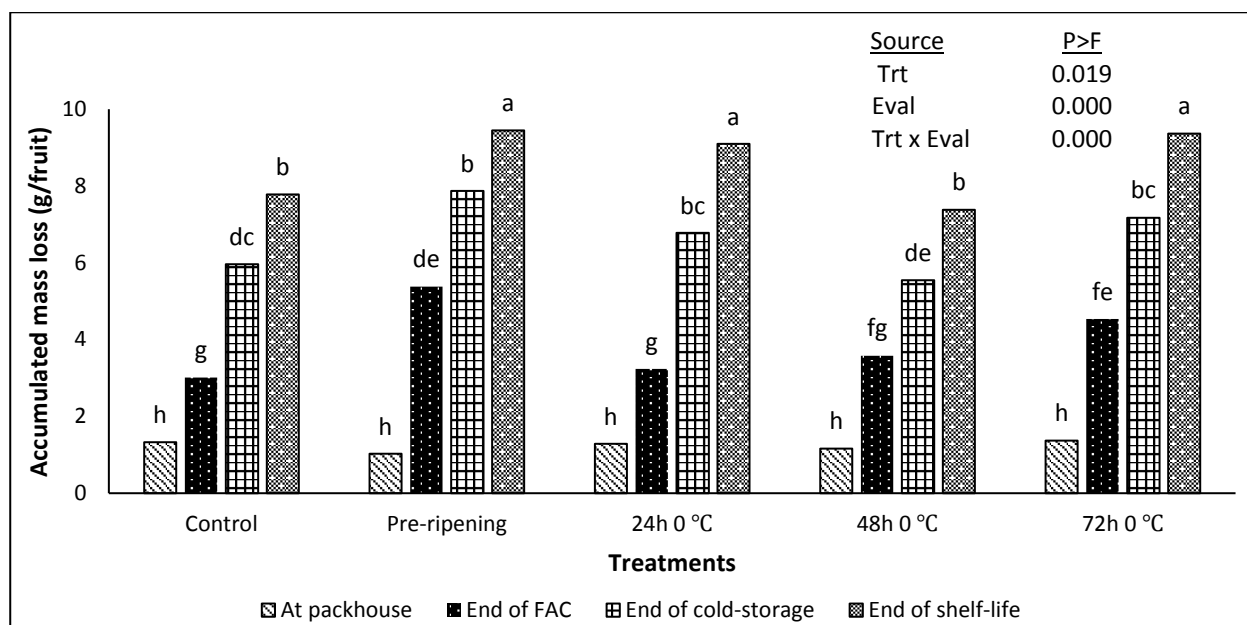


Fig. 2. Accumulated moisture loss on arrival at the pack-house, after forced air cooling (FAC), after cold-storage and at the end of shelf-life. The data of the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit mass loss. Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test. Explanation of the treatments is presented in Table 1.

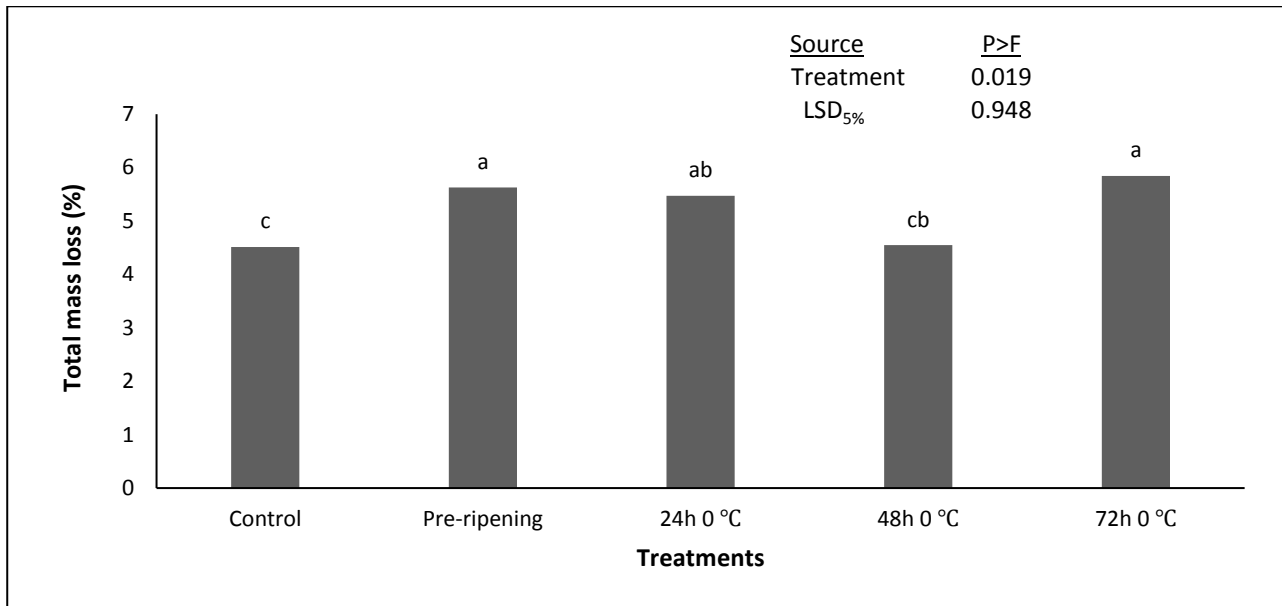


Fig. 3. Total fruit mass loss expressed as a percentage for each treatment from harvest until the end of shelf-life. The data of the two seasons were pooled to remove the variation contributed by season in order to determine the effect of treatment alone on fruit mass loss. Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test. Explanation of the treatments is presented in Table 1.

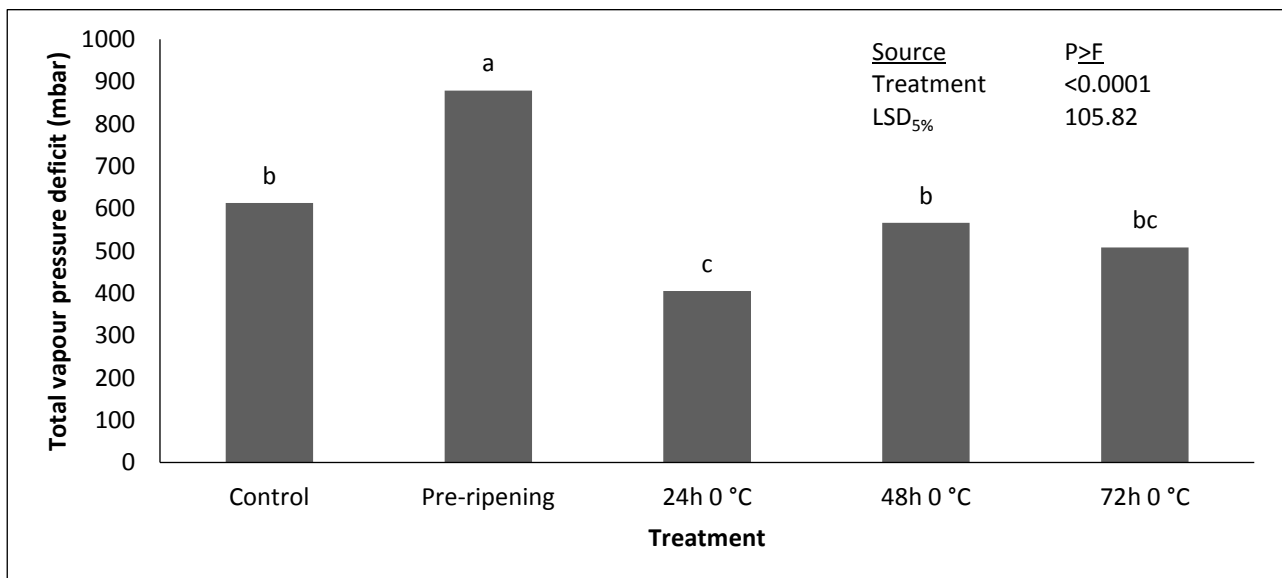


Fig. 4. Total vapour pressure deficit measured for each treatment from harvest until the end of shelf-life (2014/2015 season). Significant differences are indicated as lower case letters ($P < 0.05$) according to Fisher's LSD test. Explanation of the treatments are presented in Table 1.

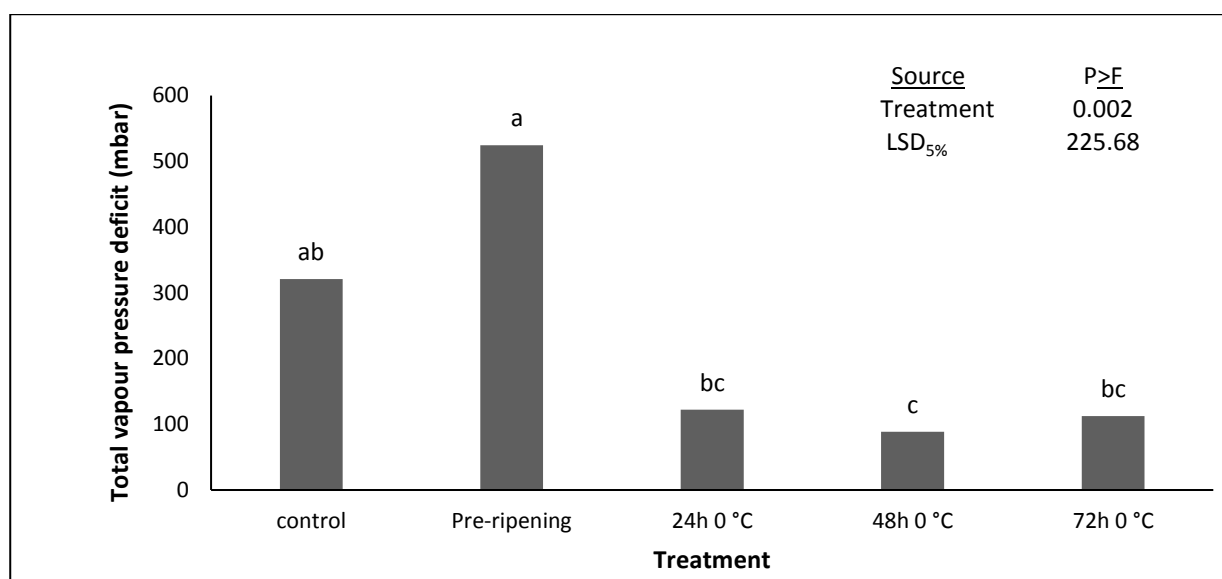


Fig. 5. Total vapour pressure deficit measured for each treatment from harvest until the end of shelf-life (2015/2016 season). Significant differences are indicated as lower case letters ($P < 0.05$) according to Fisher's LSD test. Explanation of the treatments are presented in Table 1.

PAPER 3:

The effect of pre-harvest potassium silicate application on shrivel development in 'Southern Glo' nectarine fruit

ABSTRACT

Southern Glo, an early season nectarine cultivar is characterized by relatively small fruit and hence a high surface area to volume ratio. 'Southern Glo' nectarines are prone to post-harvest shrivel and split pit. Silicon has a number of beneficial effects on fruit quality and these include reducing shrivel and split pit incidence. Therefore, it was hypothesized that through improving the strength and elasticity of cell walls, potassium silicate (K_2SiO_3) applications may reduce post-harvest shrivel and split pit incidence in 'Southern Glo' nectarines. In the 2014/2015 season three treatments were evaluated namely control, foliar application and root application (7.5 ml K_2SiO_3). In the 2015/2016 season an additional root application of 15 ml K_2SiO_3 was applied. Fruit quality was determined at harvest, after cold-storage and after shelf-life. Maturity indexing carried out in the 2014/2015 season showed the treatments did not differ significantly in their effect on total soluble solids (TSS), hue angle, flesh firmness and TA. For the 2014/2015 season the treatments differed significantly in their effect on shrivel (%) after cold storage with the root application having a significantly higher shrivel percentage (17.2 %) compared to the other two treatments. After shelf-life significant differences in flesh firmness occurred with the foliar application having a significantly higher firmness of 2.18 kg (2014/2015 season). In the 2015/2016 season the treatments did not differ significantly in any of the quality parameters measured at harvest. There were also no significant differences amongst the treatments after cold storage or shelf-life. The results obtained from both seasons indicated that the pre-harvest application of K_2SiO_3 was not effective in reducing shrivel incidence or split pit.

Keywords: 'Southern Glo', shrivel, split pit, potassium silicate, K_2SiO_3

1. Introduction

Moisture loss is a serious problem in nectarines, resulting in fruit having a shrivelled appearance and being down-graded. Fruit also lose weight during storage as a result of moisture loss, resulting in cartons not having the required weight when arriving at the market (Crisosto and Day, 2012). Southern Glo is an early season nectarine cultivar that is

susceptible to moisture loss and thus shrivel. Due to its small size it has a large surface area to volume ratio. This results in the fruit being very sensitive to moisture loss meaning that any small loss in moisture can result in serious incidence of shrivel and mass loss. Moisture loss occurs as a result of the vapour pressure deficit (VPD) between the fruit and the surrounding atmosphere (Paull, 1999). Split pit/broken stones is also a serious problem affecting the quality and marketability of stone fruit with early season cultivars more prone to split pit/broken stones compared to late season cultivars (Kritizinger, 2014).

The use of silicon containing fertilizers to improve the quality of fruit has been investigated by several researchers (Mditshwa et al., 2013; Stamatakis et al., 2003; Tarabih et al., 2014). Silicon is the second most abundant element in the earth's crust (28%) and its abundance might be the reason why it is not considered as one of the essential plant nutrients (Tefagiorgis and Laing, 2013). However, due to the widespread use of silicon containing fertilizers in Europe, silicon is now considered as a "quasi- essential" element for plant growth and development (Qiu et al., 2010). Silicon is mainly applied in the form of potassium silicate (K_2SiO_3), but other forms such as calcium ($CaSiO_3$) and sodium silicate (Na_2SiO_3) are also used.

Silicon is deposited in the plant cell walls and this helps to reinforce the cell walls by interacting with cell wall pectins and polyphenols (Stamatakis et al., 2003). This helps to protect the plant from various stresses and disease causing pathogens (Epstein, 1999; Stamatakis et al., 2003). The use of silicon containing fertilizers to reduce post-harvest weight loss in citrus fruit (Mditshwa et al., 2013) and 'Anna' apples (Mditshwa et al., 2013; Tarabih et al., 2014) has been reported. The purpose of this study was to investigate if pre-harvest K_2SiO_3 applications can maintain fruit quality post-harvest while reducing the incidence of shrivel and split pit in 'Southern Glo' nectarines.

2. Materials and Methods

2.1. Planting material

The trial was conducted on 'Southern Glo' nectarines at Welgemoed farm in Wellington, South Africa (33°35'01.8"S 18°58'53.4"E). Trees on Kakama rootstock were planted in 2003 at a spacing of 5 x 3 m. The trees were trained to an open vase system.

2.2. *Treatments and experimental layout*

In the 2014/2015 season three treatments were evaluated, namely an untreated control, foliar applications of K_2SiO_3 and root applications of K_2SiO_3 . In the 2015/2016 season an additional root application treatment of K_2SiO_3 at double rate was applied. Foliar applications were made with ten-day intervals from three weeks after full bloom until harvest. Foliar applications were done when there was no rain and the wind speed was less than 4 m s^{-1} . Root applications were applied at four-week intervals from three weeks after full bloom until harvest. AgriSil™ K50 (PQ Corporation, Wolseley), containing 33 g kg^{-1} potassium (K) and 96 g kg^{-1} silica (Si), was applied at a rate of 5 kg ha^{-1} ($7.5 \text{ ml K}_2\text{SiO}_3$ per tree) for both the foliar and root treatments in the first season. However, in the second season an additional root application treatment of 10 kg ha^{-1} ($15 \text{ ml K}_2\text{SiO}_3$ per tree) was added. For the 5 kg ha^{-1} root treatment a total of $30 \text{ ml K}_2\text{SiO}_3$ was applied per tree per season. For the additional 10 kg ha^{-1} treatment which was applied in the second season, a total of $60 \text{ ml K}_2\text{SiO}_3$ was applied per tree throughout the season. For the foliar application a total of $67.5 \text{ ml K}_2\text{SiO}_3$ was applied per tree per season. For each K_2SiO_3 foliar application, both sides of the tree were sprayed for 30 s with a motorised rucksack sprayer (Stihl, Waiblingen, Germany), delivering 2 L of solution per tree at a concentration of $100 \text{ mL } 100 \text{ L}^{-1}$ of water or 5 kg ha^{-1} . No surfactant was used for the foliar K_2SiO_3 treatments. The foliar application was done by spraying the full canopy of the trees, while the root application was done by spraying the full area under the drip line of the tree.

A randomized complete block design with ten two-tree plots per treatment was used. Two buffer trees were left between plots as well as rows where necessary to prevent carry-over of the different spray and root application treatments. Standard cultural practices were followed in the orchard.

2.3. *Data recorded*

At commercial harvest, 100 fruit were picked from each plot per block to determine incidence of fruit split and malformation. In addition, approximately 75 fruit were harvested per block per treatment to determine fruit quality at harvest (± 25 fruit) and after cold-storage (± 25 fruit) and shelf-life (± 25 fruit). The incidence of split pit and malformed fruit was determined using the Deciduous Fruit Board nectarine colour chart N.2 where fruit with a visible open split (B5 to B8) was counted. Fruit with no visible open splits at the stem-end were cut open, and if it had a split pit it was recorded. Fruit with visible malformation (A5 to

A8) were counted. Total number of split pits were calculated as the sum of the fruit with split open stem-ends plus fruit with split pit without visible signs on the outside of the fruit. Fruit were packed in 2.5 kg single layer traypack interlock cartons and wrapped with low density plastic shrivel sheets (27 micron) with 6 mm perforations for cold storage at -0.5 °C in regular atmosphere for four weeks.

At harvest the following maturity indices were recorded per block per treatment: The hue angle on both cheeks of five fruit using a calibrated colorimeter (Minolta chroma meter CR-400, Japan). Flesh firmness was determined on both peeled cheeks of ± 25 fruit using a FTA (Fruit Texture analyser, Güss Instruments) fitted with an 11 mm tip. Total soluble solids (TSS, %Brix) was determined on a pooled juice sample of ± 25 fruit using a temperature controlled, digital refractometer (Palette, PR-32 ATAGO, Bellevue, USA). Total titratable acidity (TA, %) was determined on a pooled juice sample of ± 25 fruit. TA was determined by titrating a 10 g aliquot of the juice sample with 0.1 M NaOH to a pH end-point of 8.2 using an automated titrator (Metrohm AG 760, Herisau, Switzerland).

After cold storage all plastic shrivel sheets were removed from the cartons. Cartons for the simulated shelf-life period of 7 days at 10 °C were placed at 10 °C. After cold storage and shelf-life fruit was inspected for shrivel (%) and decay (%). Shrivel was recorded when shrivelled peel extended over the shoulder of the fruit. Hue angle and flesh firmness were determined on five fruit per carton as described above. Internal defects (%) were determined by cutting the remainder of the fruit around the equatorial axis, separating the two halves of the fruit. If the fruit pulp had a dry texture with no free juice when the fruit half was squeezed, the fruit was classified as woolly. However, if the fruit had a dry texture with a little free juice running from the pulp when squeezed, the fruit was classified as pulpy. Total chilling injury (CI) was recorded as the sum of the percent woolly and pulpy fruit per replicate. Fruit was classified as overripe (OR) when abnormally soft to the touch with excessive free juice. In OR fruit the mesocarp tissue in the sub-epidermal region developed a translucent breakdown while the inner tissue exhibited a normal appearance, and/or when cut around the equatorial axis and the two halves of the fruit were twisted in opposite directions, the skin and sub-epidermal layers of the mesocarp separate from the inner mesocarp which remained attached to the stone. Total internal defects were calculated by adding CI and OR.

2.4. Data analysis

Data were analysed with two-way analysis of variance using SAS version 9.3 (SAS Institute, Inc. 2000). ANOVA-generated P-values and the significant differences between means were determined using Fisher's least significant differences (LSD) test with a 95% confidence interval when the F-statistic indicated significance at $P < 0.05$.

3. Results

3.1. Maturity indexing and split pit evaluations at harvest

At harvest in the 2014/2015 season, fruit from the different treatments did not differ significantly in flesh firmness, hue angle, TSS and TA (Table 1). However in the 2015/2016 season treatments significantly differed in TA (Table 2) with the foliar application having significantly higher TA (0.97 %) which was not significantly different from the control (0.94 %) and significantly different from the 7.5 ml root application (0.88 %) and the 15 ml root application (0.93 %). The treatments however did not have a significant effect on flesh firmness, hue angle or TSS measured at harvest in the 2015/2016 season (Table 2). 'Southern Glo' nectarines fruit at harvest did not differ significantly in the incidence of split pit at harvest in the both the 2014/2015 and 2015/2016 seasons (Tables 1 and 2).

3.2. Evaluation after cold-storage

In the 2014/2015 season fruit after cold storage did not differ significantly in flesh firmness, hue angle and decay (Table 3). However the treatments differed significantly in their effect on the shrivel (%) with the root application resulting in significantly higher shrivel percentage (17.2 %) compared to the other two treatments (Table 3). In the 2015/2016 season the treatments did not differ significantly after 4 weeks of cold storage (Table 4). The incidence of pulpiness, woolliness and over-ripeness was not recorded after cold storage in both seasons.

3.3. Evaluation after cold storage + simulated shelf-life

In the 2014/2015 season the flesh firmness was significantly higher after shelf-life following the pre-harvest foliar application of K_2SiO_3 compared to the control and the root K_2SiO_3 application treatment (Table 3). However the treatments did not have a significant

effect on the hue angle, shrivel (%) and decay (%) (Table 3). In the 2015/2016 season the treatments did not have a significant effect on any of the quality parameters measured after cold storage + simulated shelf-life i.e. flesh firmness, hue angle, shrivel (%) and decay (%) (Table 4). The incidence of pulpiness, woolliness and over-ripeness was not recorded after cold storage + simulated shelf-life in both seasons.

3.4. Mass loss

During the 2014/2015 season the treatments significantly influenced mass loss in the first phase (between harvest and end of cold storage) with the foliar K_2SiO_3 application losing significantly more mass (3.3 g fruit^{-1}) compared to the control (2.9 g fruit^{-1}) and root application (2.7 g fruit^{-1}) (Fig. 1). The treatments did not have a significant effect on the second phase of mass loss (during shelf-life). However the differences in mass loss during storage were also reflected in the accumulated mass loss over the period of cold storage and shelf-life although the foliar applied K_2SiO_3 treatment did not differ significantly from the control treatment. In the 2015/2016 season the treatments did not have a significant effect (Fig. 2) on the first phase of mass loss (between harvest and end of cold storage), but treatments had a significant effect on the second phase of mass loss (during shelf-life) with the 15 ml root application having a significantly higher mass loss (3.1 g fruit^{-1}) compared to the control (1.0 g fruit^{-1}), foliar application (1.0 g fruit^{-1}) and the 7.5 ml root application (1.0 g fruit^{-1}). The shelf-life effects were still present in the accumulated mass loss (Fig. 2).

4. Discussion

During the 2014/2015 season, the data collected at harvest showed that none of the treatments significantly affected hue angle, flesh firmness, TA and TSS. However the harvest data collected in the 2015/2016 season showed that the treatments did not significantly affect hue angle, flesh firmness and TSS, but did affect to TA. The control and the foliar K_2SiO_3 application resulted in the highest TA levels of 0.94 % and 0.97 %, respectively. Whilst the reason for the high TA content in the control is not clear, the high TA content in the foliar K_2SiO_3 may be ascribed to an increase in plant absorption of potassium, Wilkinson (2015) provided that anything causing an increase in potassium content can also result in increased fruit acidity. The firmness decreased during storage and this shows that the ripening process continued as expected during storage. A similar effect on firmness was also observed by Theron (2015) on Japanese plums. During the ripening

process the soluble pectin concentration increases as pectins are converted from the insoluble form to the soluble form, this causes fruit to soften and firmness to decrease (Von Mollendorff et al., 1993). Ethylene production during the ripening process also cause changes in the carotenoid and flavonoid content of the fruit leading to colour changes as indicated by the changing hue angle after cold storage and shelf-life compared to fruit at harvest (Giovannoni, 2004).

For the 2014/2015 season, the foliar application had a significantly higher mass loss as compared to the other treatments in the period between harvesting and the end of cold storage. This shows that the silicon applied as foliar K_2SiO_3 was not effective in reducing moisture loss during storage. Similar results were also obtained in a study on Japanese plums by Kritizinger (2014) and Theron (2015). However in the 2015/2016 season, the 15 ml root application had a significantly higher mass loss as compared to the other treatments during the shelf-life period. The results obtained showed that there was no clear relationship between mass loss and the incidence of shrivel. During the 2014/2015 season, the treatments significantly affected shrivel percentage after cold storage with the root K_2SiO_3 application increasing shrivel percentage (17.2%) compared to the other treatments. This observation was unexpected because silicon is deposited in the plant cell walls and this helps to reinforce the cell walls, protecting the plant from various stresses including moisture loss (Stamatakis et al., 2003). Mineral analysis to determine the amount of silicon in the fruit flesh were not carried out because the differences amongst the treatments were non-significant. The reason why silicon applied through the roots was not able to significantly reduce moisture loss and shrivel might be due to the fact that the applied silicon was adsorbed onto soil particles and ended up being unavailable for plant uptake (Qiu et al., 2010). However, the reason why the root application resulted in a higher shrivel percentage as compared to the control is not clear. The foliar K_2SiO_3 application did not differ from the control indicating that silicon was not effective in reducing shrivel incidence during the 2014/2015 season. The treatments did not have a significant effect on the incidence of both shrivel and decay in the 2014/2015 season. The treatments did not have a significant effect on the incidence of split pit and similar results were also obtained by Kritizinger (2014) in Japanese plums.

In the 2015/2016 season, the treatments did not significantly affect both shrivel and decay after the cold storage period and the cold storage + shelf life period. This was despite the application of an additional double rate root K_2SiO_3 application. Silicon is strongly adsorbed onto various silicon adsorption sites in the soil, i.e., mineral oxides and soil

particle-water interfaces making it unavailable for plant uptake (Qiu et al., 2010). A study by Theron (2015) to investigate the effectiveness of silicon in reducing shrivel incidence in Japanese plums also showed that silicon was not effective in reducing shrivel incidence.

5. Conclusion

The aim of this study was to determine if pre-harvest foliar and root K_2SiO_3 applications are effective in reducing split pit and post-harvest shrivel in 'Southern Glo' nectarines as silicon evidently has the ability to reduce the water vapour permeability of the fruit peel and thus reduce moisture loss and shrivel incidence. Although application of K_2SiO_3 was expected to ameliorate the moisture loss problem, this was not the case as the K_2SiO_3 only had a small effect in reducing moisture loss and shrivel. In addition, application of K_2SiO_3 did not significantly reduce the incidence of split pit in 'Southern Glo' nectarines. For future studies it is recommended to use higher rates and more frequent applications of K_2SiO_3 to determine if it can significantly reduce split pit and shrivel manifestation in 'Southern Glo' nectarines. It would also be interesting to determine silicon levels in treated fruit in order to establish whether or not significant amounts of silicon were absorbed by trees.

6. References

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Table 1.

Quality parameters measured at harvest in 'Southern Glo' nectarines. Fruit was treated with and without K_2SiO_3 (2014/2015 season).

Treatments	Quality parameters							
	Flesh firmness	Hue angle	TSS (%)	TA (%)	Fruit split (%) [*]	Split Pit (%) ^{**}	Total split pit (%) ^{***}	Malformed fruit (%) ^{****}
Control	6.16ns	19.00ns	9.48ns	0.88ns	20.0ns	7.4ns	27.4ns	14.4ns
K_2SiO_3 foliar application	6.17	20.96	9.75	0.90	27.8	3.5	31.3	17.3
K_2SiO_3 root application	6.19	19.68	10.08	0.90	25.3	5.4	30.7	15.3
LSD ($P \leq 0.05$)	1.30	2.26	0.56	0.06	7.68	3.70	8.13	12.46
Prob. > F	0.9989	0.2089	0.1108	0.6144	0.1219	0.1144	0.5657	0.8826

* According to the Deciduous Fruit Board nectarine colour chart N.2. Fruit with a visible open split (B5 to B8 on the colour chart) were counted

** Fruit with no visible open splits at the stem end were cut open, and if it had a split stone it was counted.

*** According to the Deciduous Fruit Board nectarine colour chart N.2. Fruit with visible malformation (A5 to A8 on the colour chart) were counted.

**** Total number of split stones were calculated as the sum of the fruit with split open stem ends plus fruit with split pit without visible signs on the outside of the fruit

Table 2.

Quality parameters measured at harvest in 'Southern Glo' nectarines. Fruit was treated with and without K₂SiO₃ (2015/2016 season).

Treatments	Quality parameters						
	Flesh firmness	Hue angle	TSS (%)	TA (%)	Fruit split (%) [*]	Split Pit (%) ^{**}	Malformed fruit (%) ^{***}
Control	6.33 ns	38.18 ns	11.76 ns	0.94 ab	8.00 ns	26.79 ns	9.59 ns
K ₂ SiO ₃ foliar application	6.03	40.51	11.99	0.97 a	8.94	26.40	9.50
K ₂ SiO ₃ root application (7.5ml)	6.66	37.92	11.84	0.88 c	8.45	26.99	9.41
K ₂ SiO ₃ root application (15ml)	6.20	36.95	12.13	0.93 b	8.99	27.75	9.74
LSD ($P \leq 0.05$)	1.31	3.02	0.39	0.04	5.86	5.34	4.68
Prob. > F	0.7943	0.1246	0.2492	0.0002	0.6912	0.9624	0.4089

Values in same column followed by different letters indicate significant differences ($P < 0.05$) according to Fisher's LSD test.

* According to the Deciduous Fruit Board nectarine colour chart N.2. Fruit with a visible open split (B5 to B8 on the colour chart) were counted

** Fruit with no visible open splits at the stem end were cut open, and if it had a split stone it was counted.

*** According to the Deciduous Fruit Board nectarine colour chart N.2. Fruit with visible malformation (A5 to A8 on the colour chart) were counted.

Table 3.

Quality parameters of 'Southern Glo' nectarines measured after cold-storage of 4 weeks at -0.5 °C and after cold-storage of 4 weeks at -0.5°C plus simulated shelf-life of 7 days at 10 °C. The control received no K₂SiO₃ applications, foliar K₂SiO₃ was applied every 10 days from 3 weeks after full bloom until harvest and a K₂SiO₃ root drench was applied every 4 weeks from 3 weeks after full bloom until harvest (2014/2015 season).

Quality Parameters																
Treatments		After cold-storage							After cold-storage + simulated shelf-life							
		Flesh firmness		Hue angle		Shrivel (%)		Decay (%)		Flesh firmness		Hue angle		Shrivel (%)		Decay (%)
Control	6.44	Ns	32.48	ns	7.60	b	1.20	ns	1.76	b	31.32	ns	15.20	ns	1.20	ns
K ₂ SiO ₃ foliar application	6.94		33.32		8.80	b	0.00		2.18	a	32.50		22.40		0.80	
K ₂ SiO ₃ root application	5.72		30.08		17.20	a	0.80		1.66	b	31.47		20.80		0.40	
LSD (<i>P</i> ≤ 0.05)	1.40		3.56		5.77		1.31		0.39		2.62		7.09		1.56	
<i>Prob. > F</i>	0.2127		0.1702		0.0049		0.1770		0.0317		0.5943		0.1095		0.5730	

Values in same column followed by different letters indicate significant differences (*P* < 0.05) according to Fisher's LSD test.

Table 4.

Quality parameters of 'Southern Glo' nectarines measured after cold-storage of 4 weeks at -0.5 °C and after cold-storage of 4 weeks at -0.5°C plus simulated shelf-life of 7 days at 10 °C. The control received no K₂SiO₃ applications, foliar K₂SiO₃ was applied every 10 days from 3 weeks after full bloom until harvest and K₂SiO₃ root drench applications were applied every 4 weeks from 3 weeks after full bloom until harvest (2015/2016 season).

Quality parameters								
Treatments	After cold-storage				After cold-storage + simulated shelf-life			
	Flesh firmness	Hue angle	Shrivel (%)	Decay (%)	Flesh firmness	Hue angle	Shrivel (%)	Decay (%)
Control	7.82 ns	38.89 ns	6.40 ns	1.20 ns	4.05 ns	35.54 ns	16.00 ns	1.20 ns
K ₂ SiO ₃ foliar	8.42	35.73	6.40	0.80	4.056	34.83	16.40	0.80
K ₂ SiO ₃ root (7.5ml)	8.66	36.22	5.60	1.60	3.567	35.38	14.80	0.40
K ₂ SiO ₃ root (15ml)	7.47	35.69	7.20	1.60	3.689	36.63	10.00	2.00
LSD ($P \leq 0.05$)	1.41	3.43	4.37	1.65	1.15	2.37	7.96	2.24
Prob. > F	0.3128	0.1991	0.9033	0.7179	0.7545	0.3602	0.3456	0.5037

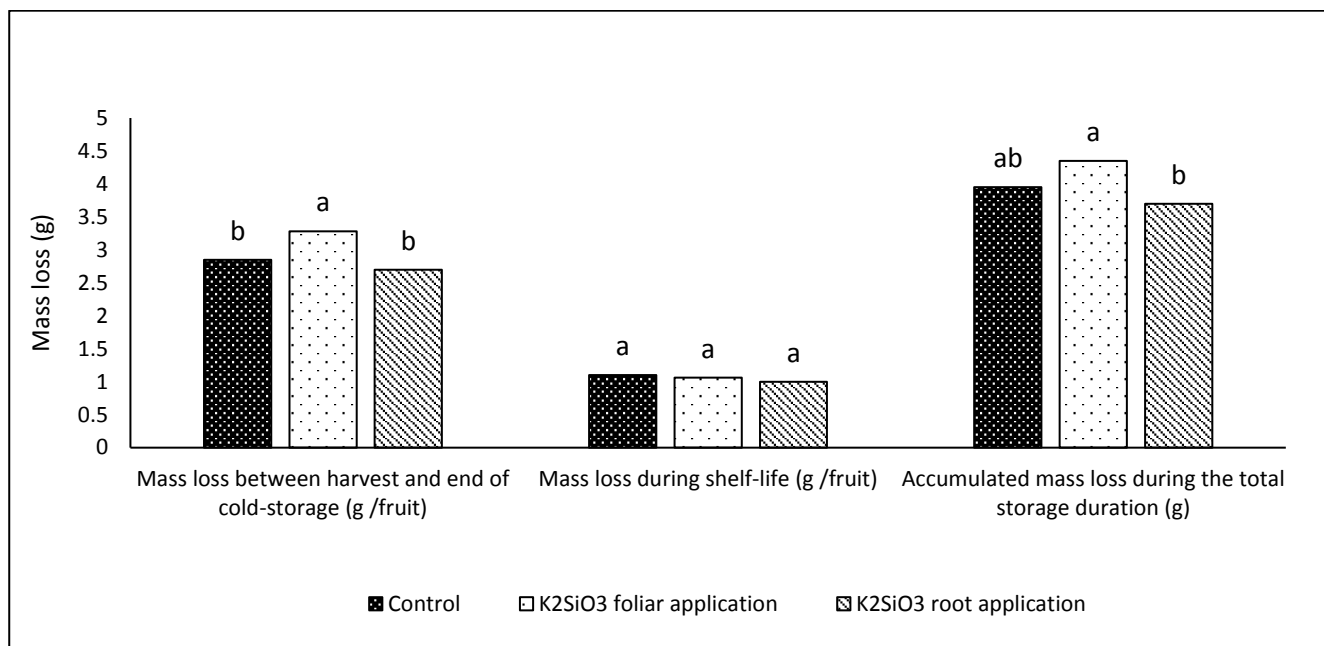


Fig. 1. Moisture loss measured in ‘Southern Glo’ nectarines at the end of cold storage and end of shelf life. The control received no K₂SiO₃ applications, foliar K₂SiO₃ was applied every 10 days from 3 weeks after full bloom until harvest and a K₂SiO₃ root drench was applied every 4 weeks from 3 weeks after full bloom until harvest (2014/2015 season).

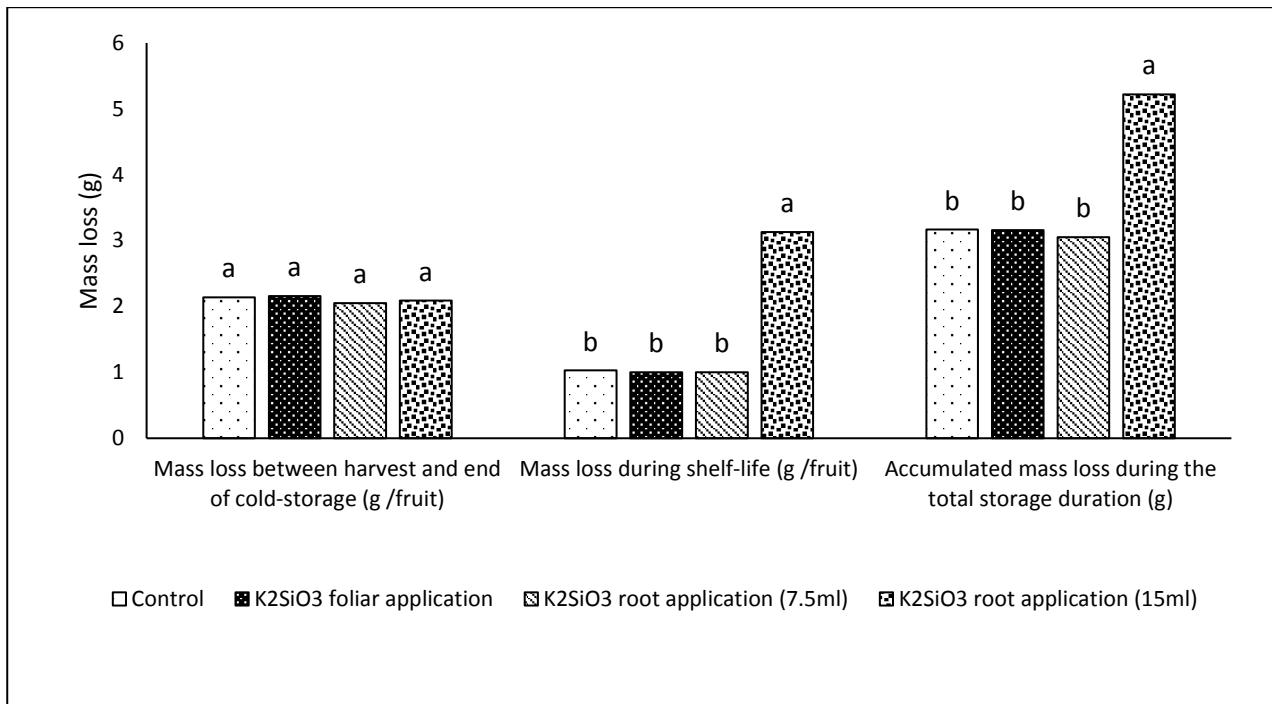


Fig. 2. Moisture loss measured in ‘Southern Glo’ nectarines at the end of cold storage and end of shelf life. The control received no K₂SiO₃ applications, foliar K₂SiO₃ was applied every 10 days from 3 weeks after full bloom until harvest and the 7.5ml and 15ml K₂SiO₃ root drenches were applied every 4 weeks from 3 weeks after full bloom until harvest (2015/2016 season).

PAPER 4:**Effect of different packaging films on moisture loss and quality of nectarines
(*Prunus persica* var. *nectarina*)**

ABSTRACT

South Africa exports approximately 3 million cartons of nectarines to overseas markets each year and the fruit usually spend four weeks in cold storage during loading, accumulation of containers and shipment to these markets. In order to minimize moisture loss and preserve fruit quality during storage, it is important that appropriate packaging materials are used. The aim of this study was to compare the effectiveness of different packaging materials in reducing moisture loss, shrivel incidence, decay and internal defects (pulpiness, woolliness and over-ripeness) in nectarines and to establish if different types of packaging material should be used for large (± 65 mm in diameter) and small (± 56 mm in diameter) nectarines. The study was carried out on two shrivel sensitive nectarine cultivars i.e. 'August Red' in the 2014/2015 season and 'Alpine' in the 2015/2016 season. A complete randomized design with 6 replicates per treatment was used. The packaging materials (treatments) used were the standard nectarine wrapper (high density polyethylene, HDPE), nectarine Xtend[®] bag, a HDPE bag with 54 x 2 mm perforations and the HDPE bag with 34 x 4 mm perforations. The same treatments were applied to both large and small fruit sizes. The fruit packed in HDPE wrappers generally had a higher percentage mass loss per fruit in both cultivars for the large and small fruit. The Xtend[®] and HDPE bags did not differ significantly in their effect on mass loss in large and small fruit of both cultivars. Shrivel was not recorded in the 2014/2015 season in either the small or large 'August Red' nectarines, but shrivel was recorded in 'Alpines' during the 2015/2016 season. For both large and small 'Alpine' nectarines, the shrivel incidence was significantly higher during evaluation after cold-storage and after shelf-life in the HDPE wrappers. The results obtained from this study show that the use of standard nectarine wrappers results in high levels of moisture loss and shrivel, while with the use of Xtend[®] or HDPE bags it was reduced. However the use of Xtend[®] or HDPE bags cannot be recommended due to the high incidence of internal defects that is associated with the use of bags.

Keywords: August Red, Alpine, moisture loss, shrivel, packaging material

1. Introduction

Nectarine (*Prunus persica* var. *nectarina*) production in South Africa mainly takes place in the Western Cape Province (HORTGRO, 2014). In the 2013/2014 season approximately 2.9 million cartons of nectarines were exported with the United Kingdom being the largest export market (54%) followed by Europe and Russia (22%) and the Middle East (19%) (HORTGRO, 2014). When exported, nectarine fruit usually spend four weeks in cold storage during loading, accumulation of containers and shipment to overseas markets and then still has a shelf-life of 5-7 days (Laubscher, 2006). 'Alpine' and 'August Red' are two of the most widely grown nectarine cultivars in South Africa and in the 2013/2014 season 684 213 and 523 309 cartons were exported, respectively (HORTGRO, 2014).

The long storage duration during shipment exposes fruit to loss of quality and in order to minimize quality loss, it is important that proper packaging materials are used. Failing this may result in weight loss, shrivel, decay and the incidence of internal defects such as woolliness, pulpiness and over-ripeness (OR) (Aharoni et al., 2007; Kaur et al., 2013; Porat et al., 2009). High density poly-ethylene (HDPE) and low density poly-ethylene (LDPE) are common types of packaging material used during fruit storage (Allahvaisi, 2012; Nath et al., 2012; Azene et al., 2014). LDPE bags or films are usually used in international transportation of fresh fruits (Scheuermann et al., 2014). LDPE is relatively inert and shrinks when heated and is a good barrier for moisture while being relatively permeable to O₂, CO₂ and volatiles (Allahvaisi, 2012). The thinner LDPE (25-38 µm) is usually used for shrink-wrapping while the thicker LDPE (45-75 µm) is used for stretch wrapping (Allahvaisi, 2012). HDPE has a higher level of crystallinity due to its non-polar and linear structure and is therefore thicker and stronger compared to LDPE (Allahvaisi, 2012; Bhunia et al., 2013) and therefore acts as a better barrier to the movement of gases and water vapour compared to LDPE (Bhunia et al., 2013). Various researchers have found that HDPE films and bags are effective in reducing moisture loss from fruit during storage (Pongener et al., 2011; Nath et al., 2012; Azene et al., 2014).

Xtend[®] films are a specialized type of modified atmosphere/modified humidity (MA/MH) packaging that have high transmission rates of water vapour (Porat et al., 2009). Compared to poly-ethylene films, Xtend[®] films have higher water vapour transmission rates (Pesis et al., 2000) and are specially designed to eliminate excess moisture that may occur inside the film (Porat et al., 2009). Because of this, the Xtend[®] films reduce moisture loss

and at the same time alleviates problems caused by water condensation inside the packaging.

The aim of this study was to compare the effectiveness of different packaging films in reducing moisture loss, shrivel incidence, decay and internal defects (pulpiness, woolliness and OR) in nectarines and to establish if different types of packaging material should be used for large (± 65 mm in diameter) and small (± 56 mm in diameter) nectarines. The packaging materials used in this study were the standard nectarine wrapper (HDPE), Nectarine Xtend[®] bag (StePac L.A. Ltd., Tefen, Israel), HDPE bag (54 x 2 mm perforations) (Peninsula Packaging, Bellville, South Africa) and the HDPE bag (34 x 4 mm perforations) (Peninsula Packaging, Bellville, South Africa). Two cultivars susceptible to shrivel were used, 'August Red' in the 2014/2015 season and 'Alpine' in the 2015/2016 season.

2. Materials and methods

2.1. Fruit sampling and experimental layout

The 'August Red' and 'Alpine' fruit were sampled from the pack-house at Timberlea farm (33°54'24.0"S 18°51'34.9"E) in Western Cape, South Africa in 2014/2015 and 2015/2016 seasons, respectively. A complete randomised design with 8 treatments and 6 replicates per treatment was used (Table 1). For treatments 1 to 4, which were large sized fruit (± 65 mm in diameter), one replicate comprised 3 cartons of fruit. One carton of fruit per replicate per treatment was used for determination of fruit maturity at harvest, and one carton each per replicate per treatment to determine fruit maturity and quality parameters after cold-storage and simulated shelf-life, respectively. For treatments 5 to 8, which comprised the smaller sized fruit (± 56 mm in diameter) packed in punnets, one replicate comprised 2 cartons of fruit. One carton of fruit per replicate per treatment was used for determination of fruit maturity at harvest, and one carton of fruit was used for the two evaluations after cold-storage and cold storage + shelf-life, respectively. As done commercially, single layer 300 x 400 mm nectarine cartons were used for the large fruit (Treatment 1 – 4) while for the smaller fruit (Treatment 5 – 8), the fruit were packed in punnets and the punnets were placed in 600 x 300 mm open display cartons. From the pack-house the fruit were transported to a laboratory at ExperiCo (PO Box 4022, Idas Valley, Stellenbosch, 7609, South Africa) for further handling and evaluation.

2.2. Data recorded

On the sampling date, maturity indexing was carried out on the fruit. Hue angle was determined with a calibrated colorimeter (Minolta chroma meter CR-400, Japan) on both cheeks of 10 fruit per replicate. Flesh firmness was determined on both peeled cheeks of 10 fruit per replicate with an FTA (Fruit Texture Analyser, Güss Instruments, Strand, South Africa), fitted with an 11 mm tip. Total soluble solids (TSS, %Brix) was determined on a pooled, juiced sample of all the fruit per replicate with a temperature-controlled, digital refractometer (Palette PR-32 ATAGO, Bellevue, USA). Total titratable acidity (TA, %) was determined on a pooled, juiced sample of all the fruit per replicate. TA was determined by titrating a 10 g aliquot of the juice sample with 0.1 N NaOH to a pH end-point of 8.2 using an automated titrator (Metrohm AG 760, Herisau, Switzerland).

For treatments 1 – 4 the weight of each fruit in the cartons designated for cold-storage and cold storage + shelf-life was recorded. For treatments 5 – 8 the weight of the fruit in each punnet was recorded in the cartons designated for cold-storage and cold storage + shelf-life. The weight of the fruit was recorded again after cold-storage and cold-storage + shelf-life, respectively. The fruit was stored at -0.5 °C for 4 weeks followed by a shelf-life period of 5 days at 10 °C.

After cold storage all packaging materials were removed from the cartons and punnets. Cartons destined for the simulated shelf-life period of 5 days were placed at 10 °C. For treatment 1 – 4 fruit from one of the two cartons per replicate was used for evaluations after cold-storage while the other carton was used for evaluations after shelf-life. For treatment 5 – 8, fruit from 3 of the 6 punnets per replicate were used for evaluations after cold storage while the other 3 punnets were used for evaluations after shelf-life. All the fruit in the carton (treatments 1 – 4) and all the fruit in 3 punnets (treatments 5 – 8) was inspected for shrivel (%) and decay (%). Shrivel was recorded when shrivelled skin extended over the shoulder of the fruit. Hue angle and flesh firmness was determined on both cheeks of 10 fruit as described above. Internal defects (%) were determined by cutting the remaining fruit per replicate around the equatorial axis, separating the two halves of the fruit. If the fruit pulp had a dry texture with no free juice when the fruit half was squeezed, the fruit was classified as woolly. However, if the fruit had a dry texture with a little free juice running from the pulp when the fruit half was squeezed, the fruit was classified as pulpy. To obtain the total chilling injury (CI), the sum of the percent woolly and pulpy fruit per replicate was calculated. Fruit was classified as OR when abnormally soft to the touch with excessive amounts of free juice, when the mesocarp tissue in the sub-epidermal region developed a

translucent breakdown while the inner tissue exhibited a normal appearance and/or when cut around the equatorial axis, and the two halves of the fruit were twisted in opposite directions, the skin and sub-epidermal layers of the mesocarp separated from the inner mesocarp which remain attached to the stone. Total internal defects were calculated by adding the total CI and the total OR.

2.3. Data analysis

Data were analysed with a mixed model repeated measures analysis of variance using Dell Inc. (2015) STATISTICA. Mass loss data were analysed using one way analysis of variance whilst the quality parameters data were analysed using two way ANOVA with packaging treatment and evaluation time being the two factors. ANOVA generated P-values and the significant differences between means were determined using Fisher's least significant differences (LSD) test with a 95% confidence interval.

3. Results

According to the maturity indexing, the harvested fruit were within prescribed export standards as required by Department of Agriculture, Forestry and Fisheries (1998) in both seasons (Table 2). Mass loss and quality parameters' data was presented separately for the large and small fruit so as to establish and show clearly the effect of the packaging treatments on the different fruit sizes and make recommendations accordingly.

3.1. Mass loss

Evaluations after cold-storage on large and small 'August Red' nectarines in the 2014/2015 season showed that the HDPE wrappers resulted in a significantly higher percentage mass loss per fruit (2% and 1.8%, respectively) compared to the other three treatments that did not differ from each other in mass loss (Fig. 1 and 2, respectively). Evaluations after cold-storage + simulated shelf-life of large and small 'August Red' nectarines confirmed that the HDPE wrappers resulted in significantly higher percentage mass loss per fruit (3.1% and 2.6%, respectively) compared to the other three treatments that still did not differ from each other (Fig. 3 and 4, respectively). During the 2015/2016 season, evaluation after cold storage showed that the HDPE wrappers resulted in significantly higher percentage mass loss per fruit in both large and small 'Alpine' nectarines

(3.2% and 3.5%, respectively) (Fig. 5 and 6, respectively). However, in the large fruit the percentage mass loss per fruit recorded in the HDPE wrappers was not significantly different from fruit in Xtend[®] bags and the Xtend bag[®] also did not differ from the HDPE bags (Fig. 5). Small 'Alpine' fruit in the Xtend[®] bags and the HDPE bags did not differ from each other in mass loss (Fig. 6). In the 2015/2016 season HDPE wrappers resulted in higher percentage mass loss per fruit (4.4%), in large 'Alpine' nectarines after cold-storage + shelf-life, but was not significantly different from the HDPE bags (54 x 2 mm perforations) (Fig. 7). The mass loss in the HDPE bags (54 x 2 mm perforations) was not significantly different from the mass loss in the Xtend[®] bags and HDPE bags (36 x 4 mm perforations), while the latter two did not differ from each other (Fig. 7). During the 2015/2016 season evaluations on small 'Alpine' nectarines after cold-storage + shelf-life showed that the percentage mass loss per fruit in the HDPE wrappers was significantly higher (5.2%) than in the Xtend[®] bags and the HDPE bags (54 x 2 mm perforations), but was not significantly different from the HDPE bags (36 x 4 mm perforations) (3.7%). The Xtend[®] bags and the HDPE bags (54 x 2 mm perforations) did not differ in mass loss percentage (Fig. 8).

3.2. Fruit quality parameters

The evaluation of the large 'August Red' nectarines in the 2014/2015 season showed no incidence of decay, shrivel and soft-tips after cold-storage for four weeks at -0.5 °C or simulated shelf-life of 7 days at 10 °C. There was no significant interaction between packaging treatment and storage duration on hue angle, flesh firmness, pulpiness, woolliness or OR (Table 3). In addition, the packaging treatment did not have a significant effect on hue angle, flesh firmness, pulpiness, woolliness and OR. However the storage duration had a significant effect on flesh firmness, pulpiness, woolliness and OR in large 'August Red' nectarines. Flesh firmness was significantly reduced during cold-storage (8.46 kg) and after shelf-life (3.78 kg). Pulpiness, woolliness and OR in large 'August Red' nectarines did not occur after cold-storage, but occurred after shelf-life (20.5%, 16.7% and 15.1%, respectively).

The evaluation of small 'August Red' nectarines during the 2014/2015 season revealed no incidence of shrivel and soft-tips after cold-storage for four weeks at -0.5 °C or simulated shelf-life of 7 days at 10 °C. Although very low levels of decay were recorded in small 'August Red' nectarines, it did not differ significantly amongst the treatments with respect to packaging treatment or evaluation date (Table 4). The packaging treatment and

storage duration did not interact significantly on flesh firmness, but flesh firmness was reduced during cold-storage to 7.57 kg and after shelf-life to only 3.29 kg (Table 4). There was a significant interaction between packaging treatment and storage duration for pulpiness of small 'August Red' nectarines. The incidence of pulpiness did not change significantly between harvest and end of cold-storage for all treatments. However, there was a general increasing trend in the incidence of pulpiness between harvest and end of shelf-life with the HDPE bags (36 x 4 mm perforations) having the highest increase in pulpiness (34.8%), the HDPE bags (54 x 2 mm perforations) had the second highest increase in pulpiness (22.9%) at the end of shelf-life and was not significantly different from the HDPE wrappers (21.6%) while the Xtend[®] had the lowest increase in pulpiness (15.6%). Packaging treatment and evaluation time did not interact significantly on woolliness in small 'August Red' nectarines. However, the storage duration had a significant effect on woolliness with the highest woolliness percentage (16.9%) being recorded after shelf-life. The interaction between packaging treatment and storage duration was significant on OR in small 'August Red' nectarines. There was no incidence of OR in all treatments between harvest and end of cold-storage. However, there was a general increase in OR between harvest and end of shelf-life, the highest increase was recorded in the Xtend[®] bags (23.8%) and HDPE wrappers (23.3%) followed by the HDPE bags (54 x 2 mm perforations) (10.4%) while the HDPE bags (36 x 4 mm perforations) did not show OR.

The evaluation of large 'Alpine' nectarines during the 2015/2016 season showed that storage duration and packaging treatment interacted significantly on flesh firmness, decay, shrivel, soft tips, pulpiness and woolliness, but not on hue angle and OR in large 'Alpine nectarines' (Table 5). Hue angle did not differ among the treatments or with evaluation time. In all the treatments, flesh firmness generally decreased from harvest until the end of cold-storage and end of shelf-life. This decrease in flesh firmness was much less in the Xtend[®] bags meaning that fruit from the Xtend[®] bags were firmer than fruit from the other three treatments at the end of shelf-life. There were generally no significant increases in the incidence of decay between harvest and end of cold-storage for the large 'Alpine' nectarines. A similar trend was followed by all treatments between harvest and end of shelf-life except for the HDPE bags (54 x 2 mm perforations) which showed a 2.9% increase in the incidence of decay. There was a general increasing trend for all treatments in the incidence of shrivel from harvest until end of cold-storage except for the Xtend[®] bag which did not show any shrivel during this period. The highest increase in shrivel incidence between harvest and end of cold-storage was recorded in the HDPE wrappers (20.8%). A similar increasing trend

was observed between harvest and end of shelf-life with the HDPE wrappers showing the highest increase in shrivel incidence (18.8%). There were generally no significant changes in the incidence of soft tips in all treatments between harvest and end of cold-storage. However, the HDPE wrappers showed a significant increase (14.5%) in the incidence of soft tips between harvest and end of shelf-life while the other three treatments did not show any significant changes during the same period. There were no significant changes in the incidence of pulpiness from harvest until the end of cold-storage in all treatments. However, there was a general increasing trend in pulpiness between harvest and end of shelf-life with the HDPE bags (36 x 4 mm perforations) showing the highest increase (13.0%) which was not significantly different from the HDPE wrappers (10.9%) while the Xtend[®] bags did not show incidence of pulpiness. The packaging treatment had a significant effect on OR with the HDPE bags (36 x 4 mm perforations) and HDPE bags (54 x 2 mm perforations) having significantly higher levels of OR (5.6% and 4.4%, respectively). The storage duration also had a significant effect on OR in large 'Alpine' nectarines with the highest OR percentage recorded between harvest and end of shelf-life (9.4%). The incidence of woolliness was not recorded in any of the treatments between harvest and end of cold-storage. However, there was a general increasing trend in the incidence of woolliness between harvest and end of shelf-life with the Xtend[®] bags having the highest increase in woolliness incidence (11.6%) while the HDPE bags (54 x 2 mm perforations) did not show incidence of woolliness.

No woolliness was recorded in evaluations of small 'Alpine' nectarines during the 2015/2016 season. Although decay and soft-tips were recorded, these did not differ significantly with respect to packaging treatment or storage duration (Table 6). Flesh firmness, shrivel and OR were significantly affected by the interaction between packaging treatment and storage duration (Table 6). In all the treatments, flesh firmness generally decreased from harvest until the end of cold-storage and end of shelf-life. This decrease in flesh firmness was much less in the Xtend[®] bags meaning that fruit from the Xtend[®] bags retained more of their firmness compared to fruit from the other 3 packaging treatments. There was a general increasing trend in the shrivel percentage between harvest and end of cold-storage and the increase was highest in the HDPE wrappers (14.4%). A similar trend was observed between harvest and end of shelf-life with the HDPE wrappers having the highest increase in shrivel percentage (21.1%). The increase in shrivel percentage in the other three packaging treatments was not significant after both cold-storage and shelf-life. The interaction between packaging treatment and storage duration did not have a significant effect on the incidence of pulpiness in small 'Alpine' nectarines, but however, the storage

duration had a significant effect on the incidence of pulpiness with the highest pulpiness percentage (1.9%) being recorded after shelf-life. There were generally no significant increases in the incidence of OR between harvest and end of cold-storage. However, there was a general increasing trend in the incidence of OR between harvest and end of shelf-life with the HDPE bags (54 x 2 mm perforations) having the highest increase in OR (16.7%) and was not significantly different from the HDPE wrappers (13.3%).

4. Discussion

The fruit packed in HDPE wrappers generally had a higher percentage mass loss compared to fruit in Xtend[®] and HDPE bags. The Xtend[®] and HDPE bags did not differ significantly in their effect on mass loss in large and small fruit for both cultivars. These findings are in agreement with Pongener et al. (2011) and Nath et al. (2012). For fruit packed in Xtend[®] and HDPE bags, a micro-atmosphere of high humidity was formed around the fruit due to the low water vapour transmission rates of Xtend[®] and HDPE bags and this reduced the vapour pressure deficit (VPD) between the fruit and the surrounding atmosphere resulting in reduced moisture loss (Azene et al., 2014; Kaur et al., 2013). The reason for the high percentage mass loss in fruit packed in HDPE wrappers could be that the wrappers allowed removal of the high humidity boundary layer around the fruit due to increased air movement compared to the Xtend[®] and HDPE bags which allowed much less air movement around the fruit (Sasthy, 1985). Azene et al. (2014) also found that papaya fruit packed in HDPE bags lost 37% less moisture in comparison to unpackaged fruit. In both cultivars, the mass loss in the large fruit was generally more than the mass loss in small fruit packed in punnets. This may be because the punnets provided an extra barrier to moisture loss resulting in less moisture being lost by the small fruit. Without the extra barrier provided by the plastic punnets, the fruit would have lost comparable amounts of moisture.

Fruit firmness decreased linearly for both cultivars in small and large fruit, the fruit were quite firm after 4 weeks of cold storage, but significant decreases in firmness were noted after the shelf-life period. The loss of fruit firmness is a physiological process that occurs during cold storage and shelf-life as a result of an increase in the soluble pectin concentration as the pectins are converted from an insoluble to a soluble form (Von Mollendorff et al., 1993). Kaur et al. (2013) also found that flesh firmness in semi-soft pear decreased linearly with increase in storage interval and they attributed this to the enzyme endopolygalacturonase which degrades soluble pectin. The results of this study showed that

the Xtend[®] bag was more effective in retaining fruit firmness compared to the other three packaging treatments, the reason for this may be that the conditions created inside the Xtend[®] bag decreased the activity of endopolygalacturonase resulting in less degradation of pectin. The slower decline in firmness of fruit packed in modified atmosphere packaging has also been documented by Kupferman and Sanderson (2005). The Xtend[®] bag is specially designed to increase the relative humidity inside the bag and this helps preserve the produce firmness (Pesis et al., 2000). Furthermore, Xtend[®] bags create a modified atmosphere inside the packaging with a high carbon dioxide and low oxygen concentration, resulting in a reduction in the ripening rate and metabolic activity inside the fruit causing the fruit to remain firmer for long (Rodov et al., 2002).

Shrivel was not recorded in the 2014/2015 season in either the small or large 'August Red' nectarines. However, shrivel was recorded in 'Alpines' during the 2015/2016 season. For both large and small 'Alpine' nectarines, the shrivel incidence was significantly higher after cold-storage and shelf-life in the HDPE wrappers and lower in the Xtend[®] and HDPE bags. This corresponds to the high percentage mass loss that was observed in the HDPE wrappers. Crouch (1998) also found a significant reduction in the incidence of shrivel in 'Laetitia' plums packed in polyethylene and polypropylene bags. The reason why fruit packed in Xtend[®] and HDPE bags had a significantly lower incidence of shrivel compared to fruit packed in HDPE wrappers may be that the bags created high humidity conditions around the fruit resulting in a low VPD between the fruit and the surrounding atmosphere thereby reducing moisture loss and shrivel (Kaur et al., 2013). Although both 'August Red' and 'Alpine' nectarines lost comparable amounts of moisture, only the 'Alpine' nectarines exhibited shrivelling whilst the 'August Red' nectarines did not show any shrivelling. The reason for this might be that 'Alpine' is generally smaller in fruit size as it is an early season cultivar and hence has a higher surface area to volume ratio. Although the aim was to use fruit of the same size in both cultivars, the 'Alpine' fruit were smaller compared to 'August Red' fruit. For example, the large 'Alpine' fruit in this study had an average diameter of ± 62 mm at harvest while the large 'August Red' fruit had an average diameter of ± 67 mm. Lownds et al. (1993) found that smaller fruit have a high surface area to volume ratio and are more sensitive to moisture loss compared to larger fruit which have a smaller surface area to volume ratio.

Internal defects (woolliness, pulpiness and OR) were recorded in both cultivars. The incidence of internal defects was generally higher after shelf-life as compared to after cold-

storage. This means that the internal defects mainly developed after fruit were transferred from cold-storage to shelf-life. Similar findings were made by Manganaris et al. (2005) who reported that pulpiness and woolliness in nectarines will only begin to develop during shelf-life after fruit is removed from cold-storage and they attributed this to an imbalance between polygalacturonase (PG) and pectin esterase (PE). OR has been attributed to increased metabolic activity and respiration rates as a result of the higher shelf-life temperature (10 °C) (Kaur et al., 2013). The incidence of internal defects was generally high in the HDPE and Xtend[®] bags for both cultivars. This was unexpected as the high humidity and carbon dioxide and low oxygen created inside the Xtend[®] bags would be expected to alleviate internal defects (Meir et al., 1997; Wang and Qi, 1997). Crouch (1998) highlighted that the use of bags can significantly reduce moisture loss and shrivel in stone fruit, but it is important not to overlook internal defect problems (pulpiness, woolliness and OR) which are associated with the use of bags. The results of this study also indicated that moisture loss and incidence of internal defects in nectarines are not directly correlated, rather, internal defects are probably a result of changes in cell wall calcium and magnesium binding properties during storage, calcium binds free water in cell walls whilst the drastic increase in cell wall magnesium content leads to impaired pectin solubilisation causing internal defects to develop (Manganaris et al., 2005; Zhou et al., 2000).

5. Conclusion

This study showed that the use of Xtend[®] and HDPE bags significantly reduced moisture loss and shrivel in nectarines in both pulp trays and plastic punnets. In both seasons the standard nectarine wrappers (HDPE wrappers) resulted in significantly high percentage mass loss per fruit. Although the Xtend[®] and HDPE bags significantly reduced moisture loss and shrivel, they also resulted in high incidence of internal defects such as woolliness, pulpiness and OR. Because of this, the use of Xtend[®] and HDPE bags cannot be recommended and the continued use of HDPE wrappers is thus recommended. The study also showed that 'Alpine' may be more susceptible to shrivel compared to 'August Red', maybe due to its smaller size and higher surface area to volume ratio which makes it more sensitive to moisture loss.

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7. Tables and Figures

Table 1.

Packaging treatments used in both the 2014/2015 and 2015/2016 seasons.

Treatment	Nectarine size*	Packaging type
1	Large	Standard nectarine wrapper
2	Large	Nectarine Xtend [®] bag
3	Large	High density polyethylene bag (54 x 2 mm perforations)
4	Large	High density polyethylene bag (34 x 4 mm perforations)
5	Small	Standard nectarine wrapper
6	Small	Nectarine Xtend [®] bag
7	Small	High density polyethylene bag (54 x 2 mm perforations)
8	Small	High density polyethylene bag (34 x 4 mm perforations)

* Large nectarine - \pm 65 mm in diameter, small nectarine - \pm 56 mm in diameter

Table 2.

Maturity of 'August Red' and 'Alpine' nectarines at harvest (standard deviations are presented in brackets).

Parameter	2014/2015	2015/2016
	August Red	Alpine
Flesh firmness (kg)	10.75 (0.96)	10.11 (0.73)
Hue angle	34.03 (4.30)	42.01 (4.60)
Total soluble solids (%)	13.19 (0.91)	11.86 (0.62)
Total titratable malic acid (%)	0.88 (0.08)	1.32 (0.07)

Table 3.

Effect of packaging treatment and cold-storage duration (evaluation date) on the quality of large (± 65 mm in diameter) 'August Red' nectarines (2014/2015 season).

Examination parameter	Evaluation ¹	Packaging treatment (A) ²						Evaluation (B) ¹						Prob. > F	
		1	2	3	4	0	1	2	A	B	A x B				
Hue Angle	0+1+2	33.24	ns	33.64	36.38	34.84	34.70	ns	33.70	35.24	0.2850	0.6559	0.0584		
Firmness (kg)	0+1+2	7.62	ns	8.25	7.87	7.84	11.28	a	8.46	b	3.74	c	0.5003	<0.0001	0.0565
Pulpiness (%)	0+1+2	6.54	ns	8.62	5.48	5.96	0.00	b	0.00	b	20.46	a	0.1197	<0.0001	0.0891
Woolliness (%)	0+1+2	6.72	ns	4.96	4.66	5.24	0.00	b	0.00	b	16.69	a	0.6993	<0.0001	0.8225
Over-ripeness (%)	0+1+2	5.80	ns	3.07	6.02	4.54	0.00	b	0.00	b	15.08	a	0.5856	<0.0001	0.7052

1 Fruit evaluations at harvest (0), after 4 weeks of cold-storage at -0.5 °C (1) and after a simulated shelf-life of 7 days at 10 °C following the cold-storage period (2).

2 Packaging treatments were as follows: 1 = HDPE wrappers with macro perforations; 2 = Xtend[®] bag; 3 = HDPE bags with 54 x 2 mm perforations; 4 = HDPE bags with 36 x 4 mm perforations.

Table 4.

Effect of packaging treatment and cold-storage duration (evaluation date) on the quality of small (± 56 mm in diameter) 'August Red' nectarines (2014/2015 season).

Examination parameter	Evaluation ¹	Packaging treatment (A) ²								Evaluation (B) ¹				Prob. > F		
		1	2	3	4	0	1	2	A	B	A x B					
Hue angle	0+1+2	33.44	ns	32.12	31.97	32.16	33.36	31.22	32.70	0.8146	0.3464	0.3976				
Firmness (kg)	0+1+2	6.66	ns	7.19	7.19	7.06	10.21	a	7.57	b	3.29	c	0.3048	<0.0001	0.6444	
Decay (%)	0+1+2	0.00	ns	0.26	0.00	0.00	0.00	ns	0.00		0.20		0.3992	0.3739	0.4338	
Pulpiness (%)	0	0.00	d	0.00	d	0.00	d						0.0350	<0.0001	0.0057	
	1	0.00	d	0.79	d	0.79	d									
	2	21.59	bc	15.62	c	22.92	b	34.84	a							
Woolliness (%)	0+1+2	3.51	ns	5.09		7.98	6.00	0.00	b	0.00	b	16.94	a	0.2099	<0.0001	0.1765
Over-ripeness (%)	0	0.00	c	0.00	c	0.00	c						0.0012	<0.0001	0.0004	
	1	0.00	c	0.00	c	0.00	c									
	2	23.30	a	23.79	a	10.42	b	0.00	c							

1 Fruit evaluations at harvest (0), after 4 weeks of cold-storage at -0.5 °C (1) and after a simulated shelf-life of 7 days at 10 °C following the cold-storage period (2).

2 Packaging treatments were as follows: 1 = HDPE wrappers with macro perforations; 2 = Xtend[®] bag; 3 = HDPE bags with 54 x 2 mm perforations; 4 = HDPE bags with 36 x 4 mm perforations.

Table 5.

Effect of packaging treatment and cold-storage duration (evaluation date) on the quality of large (± 65 mm in diameter) 'Alpine' nectarines (2015/2016 season).

Evaluation Parameter	Evaluation ¹	Packaging treatment (A) ²								Evaluation Time (B) ¹				Prob. > F				
		1		2		3		4		0		1		2		A	B	A x B
Hue Angle firmness (kg)	0+1+2	44.82	ns	42.76		42.68		44.70		44.04	ns	43.24		43.93		0.3741	0.8251	0.3385
	0	10.51	a	9.49	b	9.52	b	9.70	b							<0.0001	<0.0001	<0.0001
	1	8.15	cd	8.39	c	6.86	e	7.61	d									
	2	2.68	g	4.18	f	2.81	g	2.84	g									
Decay (%)	0	0.00	b	0.00	b	0.00	b	0.00	b							0.3896	0.0404	0.0041
	1	1.45	ab	0.73	b	0.00	b	0.73	b									
	2	0.00	b	0.73	b	2.90	a	0.00	b									
Shrivel (%)	0	0.00	e	0.00	e	0.00	e	0.00	e							<0.0001	<0.0001	<0.0001
	1	20.77	a	0.00	e	15.32	bc	6.52	d									
	2	18.84	ab	6.52	d	7.97	d	10.87	cd									
Soft tips (%)	0	0.00	b	0.00	b	0.00	b	0.00	b							<0.0001	<0.0001	<0.0001
	1	0.00	b	0.00	b	0.00	b	1.45	b									
	2	14.50	a	1.45	b	0.00	b	0.00	b									
Pulpiness (%)	0	0.00	d	0.00	d	0.00	d	0.00	d							<0.0001	<0.0001	0.0127
	1	1.39	cd	0.00	d	0.00	d	0.00	d									
	2	10.87	ab	0.00	d	6.52	bc	13.04	a									
Overripeness (%)	0+1+2	2.17	b	2.17	b	4.35	ab	5.56	a	0.00	b	1.27	b	9.42	a	<0.0001	<0.0001	0.0572
Woolliness (%)	0	0.00	b	0.00	b	0.00	b	0.00	b							0.0000	0.0000	0.0021
	1	0.00	b	0.00	b	0.00	b	0.00	b									
	2	0.73	b	11.59	a	0.00	b	2.17	b									

1 Fruit evaluations at harvest (0), after 4 weeks of cold-storage at -0.5 °C (1) and after a simulated shelf-life of 7 days at 10 °C following the cold-storage period (2).

2 Packaging treatments were as follows: 1 = HDPE wrappers with macro perforations; 2 = Xtend[®] bag; 3 = HDPE bags with 54 x 2 mm perforations; 4 = HDPE bags with 36 x 4 mm perforations

Table 6.

Effect of packaging treatment and cold-storage duration (evaluation date) on the quality of small (± 56 mm in diameter) 'Alpine' nectarines (2015/2016 season).

Evaluation Parameter	Evaluation ¹	Packaging treatment (A) ²								Evaluation Time (B) ¹				Prob. > F		
		5	6	7	8	0	1	2	A	B	A x B					
Hue Angle firmness (kg)	0+1+2	36.52	b	41.53	a	40.24	a	40.62	a	39.97	ns	39.75	39.46	0.0007	0.8917	0.2477
	0	10.18	a	10.45	a	10.67	a	10.35	a					0.0002	<0.0001	0.0177
	1	8.67	c	9.40	b	8.63	c	9.29	bc							
	2	2.97	e	4.67	d	3.42	e	3.48	e							
Decay (%) Shrivel (%)	0+1+2	0.00	ns	0.00		0.00		0.74		0.00	ns	0.28	0.28	0.1236	0.6090	0.8059
	0	0.00	b	0.00	b	0.00	b	0.00	b					<0.0001	0.0002	0.0215
	1	14.44	a	0.00	b	3.33	b	5.56	b							
	2	21.11	a	5.56	b	3.33	b	4.44	b							
Soft tips (%)	0+1+2	1.11	ns	0.37		0.00		0.00		0.00	ns	0.28	0.83	0.2138	0.2680	0.0645
Pulpiness (%)	0+1+2	1.11	ns	1.48		0.00		0.00		0.00	b	0.00	b	1.94	a	0.2803
Overripeness (%)	0	0.00	c	0.00	c	0.00	c	0.00	c					0.0887	<0.0001	0.0055
	1	0.00	c	1.11	c	0.00	c	1.11	c							
	2	13.33	a	6.67	b	16.67	a	7.78	b							

1 Fruit evaluations at harvest (0), after 4 weeks of cold-storage at -0.5 °C (1) and after a simulated shelf-life of 7 days at 10 °C following the cold-storage period (2).

2 Packaging treatments were as follows: 1 = HDPE wrappers with macro perforations; 2 = Xtend[®] bag; 3 = HDPE bags with 54 x 2 mm perforations; 4 = HDPE bags with 36 x 4 mm perforations

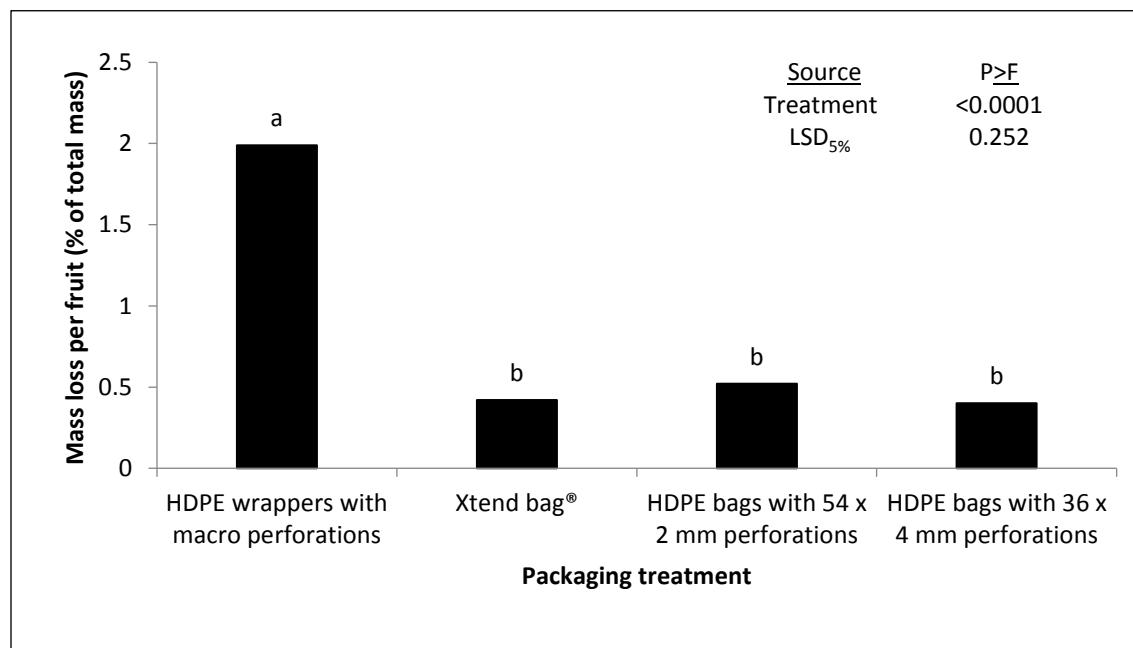


Fig. 1. Percentage mass loss per fruit in large (± 65 mm in diameter) 'August Red' fruit during cold storage. Fruit was packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C (2014/2015 season).

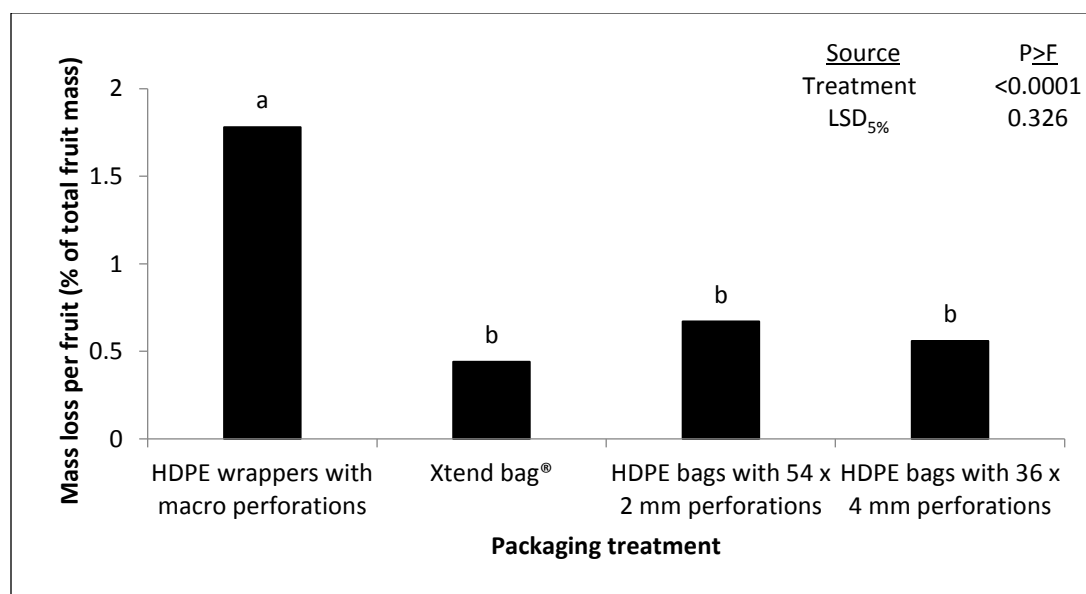


Fig. 2. Percentage mass loss per fruit in small (± 56 mm in diameter) 'August Red' fruit during cold storage. Fruit was packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C (2014/2015 season).

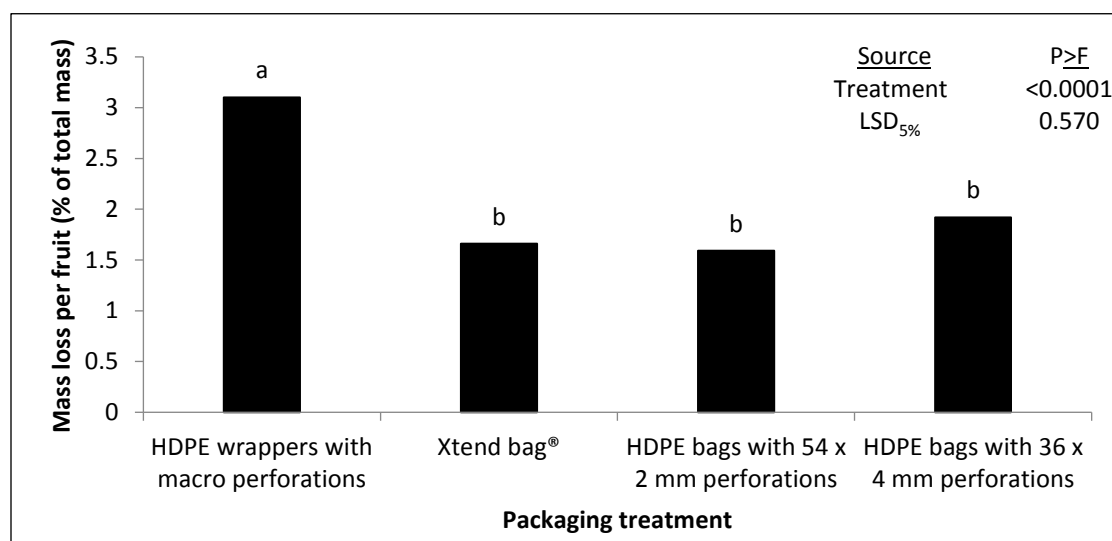


Fig. 3. Percentage mass loss per fruit in large 'August Red' fruit (± 65 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C plus a simulated shelf-life of 5 days at 10 °C (2014/2015 season).

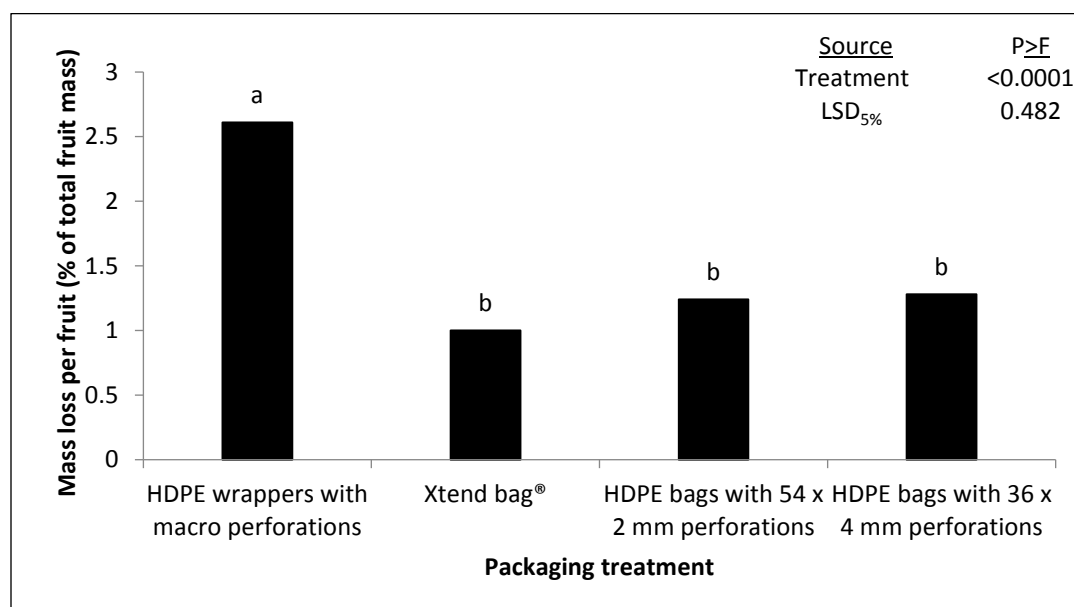


Fig. 4. Percentage mass loss per fruit in small 'August Red' fruit (± 56 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C and 5 days of simulated shelf-life at 10 °C (2014/2015 season).

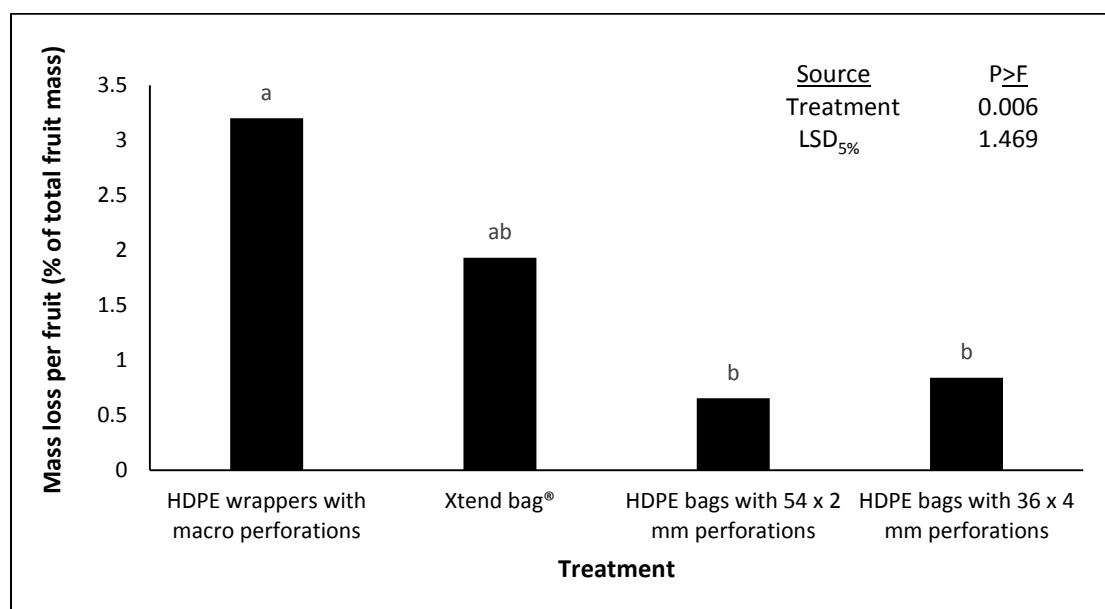


Fig. 5. Percentage mass loss per fruit in large 'Alpine' fruit (± 65 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C (2015/2016 season).

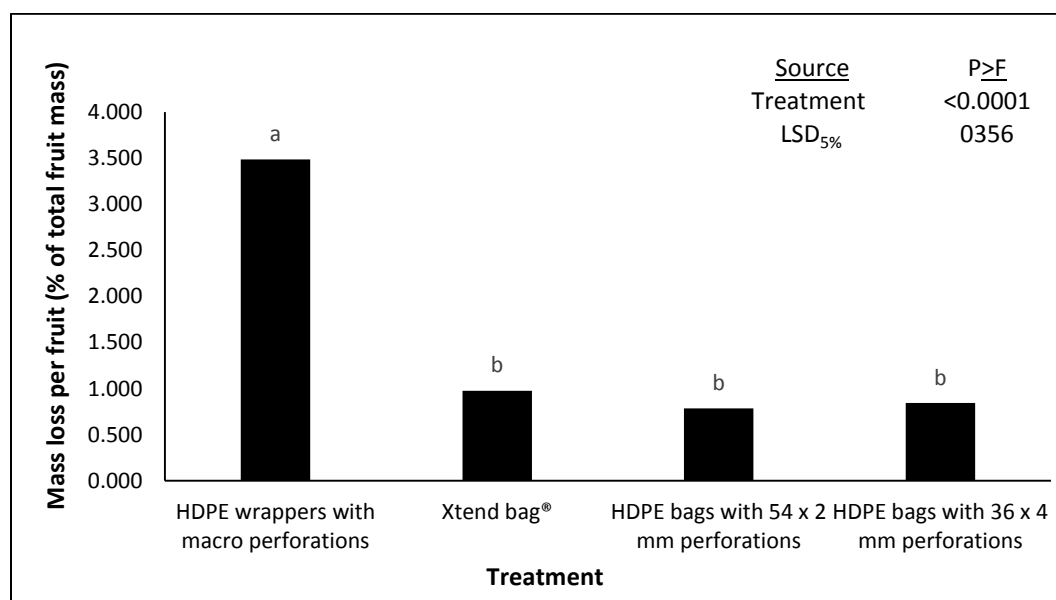


Fig. 6. Percentage mass loss per fruit in small 'Alpine' fruit (± 56 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C (2015/2016 season).



Fig. 7. Percentage mass loss per fruit in large 'Alpine' fruit (± 65 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C plus a simulated shelf-life of 5 days at 10 °C (2015/2016 season).

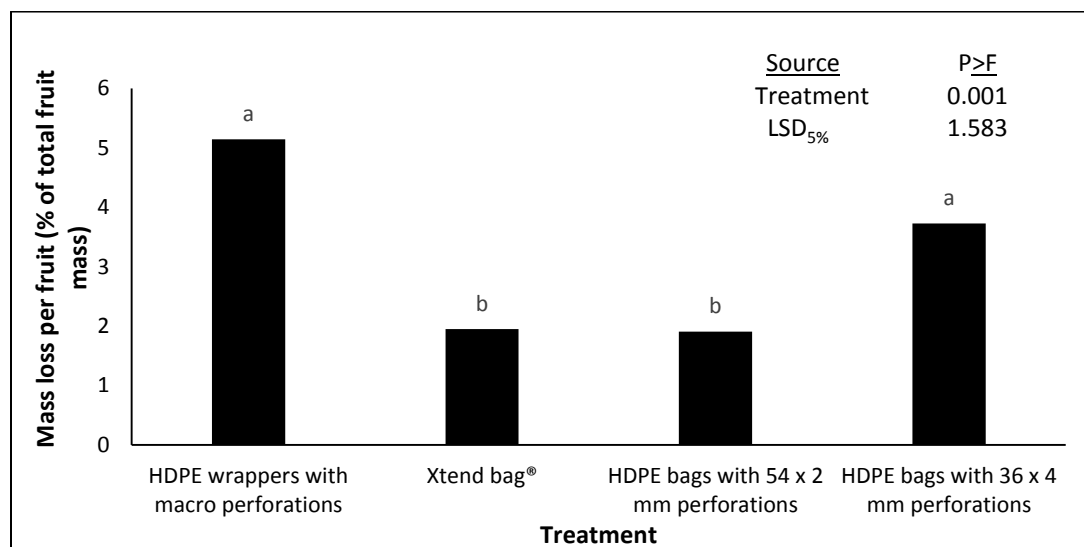


Fig. 8. Percentage mass loss per fruit in small 'Alpine' fruit (± 56 mm in diameter) packed in perforated high density polyethylene wrappers, Xtend® bags, high density polyethylene bags with 54 x 2 mm perforations and high density polyethylene bags with 34 x 4 mm perforations and stored 4 weeks in cold storage at -0.5 °C and 5 days of simulated shelf-life at 10 °C (2015/2016 season).

GENERAL DISCUSSION AND CONCLUSION

Moisture loss is one of the main post-harvest problems that affect the quality of peaches and nectarines during long term storage (Crisosto and Day, 2012). To ensure optimum post-harvest life, stone fruit such as peaches and nectarines should be protected from excessive post-harvest moisture loss (Crisosto and Day, 2012). According to Holcroft (2015) peaches and nectarines have a water content of about 89% and will show symptoms of shrivel when they have lost at least 19% of this water. Moisture loss during long term storage can result in fruit with a shrivelled appearance rendering them unsaleable (Maguire et al., 2000). In addition to this, moisture loss results in loss of saleable weight as well as deterioration of fruit quality (Sastri, 1985). Moisture is lost from the fruit as a result of the vapour pressure deficit (VPD) between the fruit and the surrounding atmosphere. Fruit moisture loss occurs through various parts of the fruit surface, these include the stomata, lenticels, cuticle, and epicuticular wax platelets (Díaz-Pérez et al., 2007). Moisture loss accounts for over 97% of the total weight loss in harvested produce (Díaz-Pérez et al., 2007).

In Paper 1 we report on the effect of fruit to fruit variation, harvest date, tree and orchard effects and cultivar differences on water vapour permeance ($P'H_2O$) of three nectarine cultivars namely 'Alpine' (susceptible to post-storage shrivel), 'Summer Bright' (not susceptible to post-storage shrivel) and 'August Red' (highly susceptible to post-storage shrivel). The study showed that the peel of 'Summer Bright' nectarines was more permeable to water vapour compared to 'August Red' and 'Alpine'. This is despite the fact that 'Summer Bright' is not susceptible to post-storage shrivel, therefore $P'H_2O$ does not appear to be an accurate predictor whether a cultivar is susceptible to shrivel or not. The study also showed that large fruit to fruit variation were the main contributor (>45%) to the variation in $P'H_2O$ within a population of harvested fruit. The large fruit to fruit variation make it difficult to effectively manage the fruit so as to reduce moisture loss and shrivel incidence. However, there is need for further research to establish the extent to which other factors such as position of fruit within canopy, fruit contact with stems and leaves, exposure to sunlight, and size and shape of the fruit influence the fruit $P'H_2O$. Harvesting date made the second largest contribution (> 35%) to the total variance overall and in

each of the three cultivars. In both seasons it was observed that there was generally an increase in $P'H_2O$ in 'Alpines', 'Summer Bright' and 'August Red' nectarines with later sampling dates. Similar findings were made by Maguire et al. (2000) in apple and Theron (2015) in Japanese plums. With this in mind, it is important to implement measures to reduce moisture loss during harvesting. Such measures include harvesting fruit in the cooler time of the day, keeping fruit under a shade after harvesting and covering fruit with wet blankets.

Cultivar differences explained more than 7% of the total variation in $P'H_2O$ of the three nectarine cultivars. Variation in $P'H_2O$ amongst different cultivars may be caused by variation in the occurrence of micro-cracks and differences in soluble cuticular lipids and their molecular structure (Lescourret et al., 2001). In addition to differences in the occurrence of micro-cracks amongst different cultivars, variation in $P'H_2O$ amongst cultivars is also related to differences in the physical and chemical properties of outer layers of the fruit, cuticle thickness as well as amount and type of epicuticular waxes (Maguire et al., 2000). Orchard effects explained only 3% of the total variation in $P'H_2O$ for both seasons overall and in each of the three cultivars. This shows that the orchards were relatively uniform and therefore did not make a big contribution to the variation in $P'H_2O$. Although the differences amongst the orchards were small, these differences may have been due to different orchard management practices. Tree differences did not contribute to the total variation in $P'H_2O$. The reason for this might be that $P'H_2O$ is a fruit characteristic and influences from the whole tree physiology are very small (Maguire et al., 2000).

In Paper 2, we determined the VPD between 'August Red' nectarines and their environment during different simulated post-harvest handling chains. The fruit were subjected to different handling chains from harvest until end of shelf-life so as to determine the moisture loss at different stages in the handling chains. Currently the handling protocol for nectarines in South Africa is to harvest fruit during the cooler time of the day (temperature under 25 °C) and to remove field heat as soon as possible after harvesting by cooling fruit to just above the dew point temperature of the pack-house (HORTGRO, 2014). Furthermore, the handling protocol requires that nectarines should

be packed within 12 hours of arrival at the pack-house (HORTGRO, 2014). The use of pre-ripening to reduce incidence of pulpiness and woolliness has been reported by several researchers (Laubscher, 2006; Lurie and Crisosto, 2005; Nanos and Mitchell, 1991) and in this study we added a pre-ripening stage to one of the treatments to see if pre-ripening can indeed reduce internal breakdown without increasing shrivelling in 'August Red' nectarines. Shrivel was not recorded in any of the handling chains after both cold-storage and cold storage + shelf-life, as the amount of moisture lost was not high enough to induce shrivel in 'August Red' nectarines. According to Holcroft (2015) peaches and nectarines have a water content of about 89% and will only show symptoms of shrivel when they have lost at least 19% of this water. In our study the nectarines only lost an average of 5% of mass and this was therefore not sufficient to induce shrivel. From the results of this study it was clear that none of the proposed handling chains performed better than the control (recommended industry handling protocol) in terms of reducing moisture loss, shrivel and internal breakdown. It is therefore recommended that the industry handling protocol should be carefully followed so that moisture loss and its associated problems are kept to a minimum.

The aim in Paper 3 was to investigate if pre-harvest potassium silicate (K_2SiO_3) applications can maintain fruit quality post-harvest while reducing the incidence of shrivel and split pit in 'Southern Glo' nectarines. The use of silicon containing fertilizers to improve the quality of fruit has been investigated by several researchers (Mditshwa et al., 2013; Stamatakis et al., 2003; Tarabih et al., 2014). According to Qiu et al. (2010), silicon applied through the soil may become adsorbed to soil particles limiting its availability for plant uptake. With this in mind, an additional foliar application treatment was also done to see if it can help in reducing moisture loss. Although application of K_2SiO_3 was expected to ameliorate the moisture loss problem, this was not the case as the K_2SiO_3 only had an insignificant effect in reducing moisture loss and shrivel. In addition to this, application of K_2SiO_3 did not significantly reduce the incidence of split pit in 'Southern Glo' nectarines. For future studies it is recommended to use higher rates and more frequent applications of K_2SiO_3 to determine if it can significantly reduce split pit and shrivel manifestation in 'Southern Glo' nectarines.

Lastly in Paper 4 we compared the effectiveness of different packaging materials in reducing moisture loss, shrivel incidence, decay and internal defects (pulpiness, woolliness and over-ripeness (OR) in nectarines and established if different types of packaging material should be used for large (68-74 mm in diameter) and small (59-61 mm in diameter) nectarines. The long storage duration during shipment to overseas markets exposes fruit to loss of quality and to combat this, it is important that proper packaging materials are used. Failure to package fruit properly may result in weight loss, shrivel, decay and the incidence of internal defects such as woolliness, pulpiness and OR (Aharoni et al., 2007; Kaur et al., 2013; Porat et al., 2009). The results of this study showed that the use of Xtend® and HDPE bags significantly reduced moisture loss and shrivel in nectarines in both pulp trays and plastic punnets. During both seasons of this study, the standard nectarine wrappers (HDPE wrappers) resulted in significantly more mass loss per fruit. However, the use of Xtend® and HDPE bags resulted in a significantly high incidence of internal defects such as woolliness, pulpiness and OR. The use of bags is therefore not recommended for both the large fruit packed in pulp trays and small fruit packed in punnets. The use of HDPE wrappers is thus recommended.

Overall, this study found that the peel permeability of nectarines is high at optimum harvest and measures to reduce moisture loss should be implemented during harvesting. In addition to this, the study also highlighted that the current standard handling protocol for nectarines in South Africa should be carefully followed and unnecessary delays in cooling of fruit after harvest should be avoided. Furthermore the study also showed that it is important to limit the time during which fruit is exposed to high pre-ripening temperatures so that moisture loss is kept to a minimum. In order to maintain the quality of fruit at acceptable levels when they get to their markets, it is advisable to use Xtend® and HDPE bags instead of HDPE wrappers. This helps to reduce the VPD between fruit and the surrounding atmosphere thereby alleviating the moisture loss problem.

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